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A ruminant animal feed comprising a forage component with about 20% to about 100% corn silage produced from corn plants exhibiting a *bm* phenotype and a feed composition component and a method of producing the same is disclosed. A method of enhancing milk production in ruminant animals, especially dairy cattle, by feeding them with the animal feed of the invention, optionally co-administering somatotropin, is also disclosed.

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(54) Animal feed

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**Description**

This invention relates to increasing milk production in ruminant animals such as dairy cows. More particularly the invention relates to increasing milk production in animals such as dairy cows by feeding a ration comprising silage from 5 corn plants exhibiting a brown midrib phenotype.

Corn plants (*Zea mays L.*) are bred by both self-pollination and cross-pollination techniques. Corn is a monoecious plant, i.e., each plant has separate male and female flowers on the same plant, located on the tassel and ear, respectively. Natural pollination occurs in corn when pollen is shed from tassels and contacts silks of the same plant or a different plant that protrude from tops of the developing ears. Methods and techniques for the development of inbred 10 corn lines and hybrid corn varieties are known in the art. Hallauer, A., *Maize*, in *Principles of Cultivar Development*, Vol. 2, Fehr, W. ed. pp. 249-294, Macmillan, New York, (1987). Currently, many hybrid corn varieties are produced by crossing two inbred lines to product  $F_1$  hybrid progeny. The  $F_1$  plants exhibit heterosis, or hybrid vigor, resulting in plants having high yield and superior agronomic performance in the hybrid combination. The production and development of inbred corn lines and hybrid corn varieties are discussed in, for example, U.S. Patent Nos. 5,367,109 and 15 5,495,067, which are incorporated herein by reference.

Research studies on maize have resulted in the identification of numerous genetic loci. See, e.g., the maize genetic database on the Worldwide Web at <http://teosinte.agron.missouri.edu/top.html>.

To supply needed nutrients for increased milk production by lactating dairy cattle is an ongoing challenge facing the dairy industry. This challenge is complicated by the fact that, even though a dairy cow's diet may meet the National 20 Research Council recommended nutrient requirements, the diet may still lack some nutrients at increased levels required for higher milk production. One reason for this difficulty in meeting nutrient requirements is the complexity of the digestive system of ruminants such as dairy cattle.

In cattle, ingested feed first passes into the reticulorumen, where it is subject to anaerobic microbial fermentation. Microbial fermentation begins the digestive process and gives a ruminant the ability to utilize fibrous feeds, in contrast 25 to monogastric animals. Ruminants meet their nutrient needs by utilizing the by-products of microbial fermentation, along with any undigested feed residues and the resultant microbial mass that passes from the rumen.

Anaerobic microbial fermentation is an advantage to ruminants because it allows them to benefit from feeds which cannot be utilized by non-ruminants. However, microbial activity limits the ability to provide supplemental nutrients to a ruminant animal, because many desirable nutrients, such as proteins, amino acids and digestible fiber, will be metabolized by microbes before the nutrients reach a site where they can be absorbed and utilized by the ruminant.

Attempts have been made to increase milk production in dairy cattle by manipulating the feed ration. For example, rations containing silage derived from corn plants carrying a brown midrib (*bm*) mutation have been fed to cattle. Stallings, C. et al., *J. Dairy Sci.*, 65:1945-1949 (1982); Block, E. et al., *J. Dairy Sci.* 64:1813-1825 (1981); Keith, E. et al., *J. Dairy Sci.* 62:788 (1979). The *bm* gene decreases and alters the lignin content in the vegetative parts of such 35 corn plants; silage made from such plants has increased fiber digestibility compared to silage from corn plants not exhibiting the brown midrib phenotype. In general, these studies indicated that there was no increase in milk production in cows fed silage from *bm* corn. It was concluded that the cows fed a diet containing *bm* silage generally partitioned the nutrients into meat or fat body tissues rather than milk production. Barriere et al., *Agronomie* 13:865-876 (1993).

Attempts have been made to increase the efficiency of feed utilization and milk production by using various formulations and feed supplements. Despite continued improvement in the development of dairy cattle feed rations, it is desirable to further increase the efficiency of feed utilization and milk production by dairy cattle.

It has now surprisingly been found that silage from *bm* corn may be used to increase the milk production of ruminant animals when combined to make an animal feed with defined constituents.

Thus, viewed from one aspect the present invention provides a ruminant animal feed comprising a combination of:

45 a forage component comprising from about 20% to about 60% of said feed on a dry matter basis, said forage component comprising from about 20% to about 100% corn silage on a dry matter basis produced from corn plants exhibiting a brown midrib (*bm*) phenotype, said silage having an *in vitro* neutral detergent fibre digestibility of about 44% to about 70%; and

50 a feed composition component;  
said animal feed having a fibre content of about 20% to about 40%. Preferably the animal feed of the invention is in the form of a total feed ration, preferably a total dairy cattle feed ration.

The ration can have a crude protein content of from about 17% to about 21% on a dry matter basis. About 30% 55 to about 50% of the crude protein is soluble protein.

The neutral detergent fiber digestibility can be from about 6% to about 20% greater than the neutral detergent fiber digestibility of corn silage produced from corresponding isogenic normal corn plants. The silage can have a whole plant *in vitro* digestibility from about 65% to about 85%.

The corn plants can comprise  $F_1$  hybrid plants. The brown midrib phenotype can be the result of homozygosity at the *bm3* gene locus.

Viewed from a further aspect the invention provides a method of enhancing milk production in a ruminant animal, such as cattle, sheep or goats, preferably in a dairy cow, comprising the step of feeding said animal an animal feed as defined hereinbefore. Alternatively viewed the present invention provides an animal feed as described herein for enhancing milk production in a ruminant animal. Alternatively, the use of a forage component or a feed composition component as described herein for the preparation of an animal feed as described herein for enhancing milk production in a ruminant animal, is also provided.

The method can further comprise the step of administering a biologically active somatotropin to the animal, e.g. a cow under conditions delivering an effective amount of the somatotropin to the animal during a selected period. The somatotropin can be administered as a prolonged release dose, for example, a dose that is effective for at least 7 days. The conditions can comprise delivering the somatotropin to the circulatory system of the animal.

In some embodiments, the forage component comprises from about 40% to about 60% of the ration, and the *bm* corn silage comprises from about 40% to about 80% of the forage component. In some embodiments, the forage component comprises from about 45% to about 55% of the ration and the *bm* corn silage comprises from about 50% to about 80% of the forage component.

The present invention further extends to a method of producing a total ruminant animal feed as defined herein comprising the steps of a) obtaining a corn silage produced from corn plants exhibiting a *bm* phenotype; and b) formulating a total ruminant animal feed comprising a combination of a forage component and a feed composition component as defined herein.

Preferably in this method said corn plants are grown from a substantially homogenous assemblage of corn seeds which are homozygous for at least one *bm* allele.

According to a yet further aspect of the invention we provide a pack for a ruminant animal feed comprising a forage component and separately a feed composition component as defined herein as a combined preparation for simultaneous, separate or sequential use to enhance milk production in said animal. Such a pack may additionally comprise a biologically active somatotropin. Novel plants and seeds described herein also form aspects of the invention. The seeds of the invention may be viable or alternatively non-viable, such as crushed, milled or pressed seeds. Silage produced from the novel seeds and plants of the invention form yet further aspects of the invention.

An article of manufacture is disclosed, comprising packaging material, a substantially homogenous assemblage of  $F_1$  hybrid corn seeds within the packaging material and a package label accompanying the packaging material. The seeds can be homozygous for *bm3*. The package label indicates that the seeds are effective for producing silage that increases milk production when fed to lactating dairy cattle.

The package label can indicate that the seeds are effective for producing silage having an *in vitro* neutral detergent fiber digestibility of from about 44% to about 70% and a whole plant *in vitro* digestibility of about 65% to about 85% determined after ensiling the plants.

The package label can also indicate that the seeds are effective for producing silage that increases milk production when fed to lactating dairy cows in a ration having a fiber content of about 20% to about 40%. Such a ration comprises about 20% to about 60% of a forage component, with the remainder being a feed composition. The forage component comprises from about 20% to about 100% corn silage produced from corn plants homozygous for *bm3*. The package label can also indicate that the ration has a crude protein content of from about 17% to about 21% on a dry matter basis and about 35% to about 50% of the crude protein is soluble protein.

Figure 1 shows herd milk production on a daily basis in a switchback trial in which normal or BMR silage was fed in conjunction with bovine somatotropin administration.

A feed ration has been discovered that comprises a corn silage produced from corn plants exhibiting a brown midrib phenotype. Feeding of such silage to dairy cattle results in unexpected improvements in milk production.

A silage component of the feed ration is produced from corn plants displaying a brown midrib (*bm*) phenotype. The *bm* phenotype is exhibited by plants homozygous for a mutant allele at the *bm1*, *bm2*, *bm3* or *bm4* loci. In some embodiments, such plants may display the brown midrib phenotype due to homozygosity at more than one of the *bm* loci. Mutant *bm* alleles are known to reduce and alter the lignin content in plants homozygous for such alleles. The lignin content may be reduced 20%, 30%, or up to about 45% compared to corn of the same genetic background but having a wild-type *Bm* gene.

Corn inbreds and hybrids carrying *bm* alleles and displaying the *bm* phenotype can be produced by corn breeding methods such as conversion programs or recurrent selection. In one embodiment, a corn inbred line is converted to the *bm* phenotype in a breeding program initiated from the  $F_1$  progeny of a cross between a plant of a first inbred (wild-type for the *bm* phenotype) and plants of a second line carrying the desired *bm* allele.  $F_1$  plants are backcrossed to the first inbred line until an inbred line is obtained that has substantially the same genotype as the original inbred line except for the replacement of the wild-type *Bm* gene by the mutant *bm* gene. Conversion programs, recurrent selection programs, pedigree breeding programs, breeding programs using synthetics and other breeding methods are described

in, e.g., Hallauer, et al. in *Corn and Corn Improvement*, Sprague et al., eds. pp. 463-564 (1988).

In addition to selecting and identifying plants containing a mutant *bm* gene, it is desirable to select concomitantly for plants having superior agronomic and yield performance characteristics.

Techniques for identifying plants displaying the brown midrib phenotype are known in the art. For example, the underside of leaves may be examined at 10-14 days before tassel emergence (4-6 leaf stage, 0.6-1 meter height) for the appearance of a golden-brown or reddish-brown color on the midrib. Plants may also be examined at maturity by removing a leaf sheath and examining the stalk. The stalk has a golden-brown or reddish-brown color if the brown midrib phenotype is expressed. Brown pigment is also present in the cob and in the roots. Because the *bm* phenotype is recessive, the presence of the *bm* gene in heterozygotes can be determined by performing a self and evaluating the selfed progeny for the expected 3:1 segregation ratio. Alternatively, marker-assisted breeding techniques may be used, e.g., restriction fragment length polymorphisms (RFLP), simple sequence repeats (SSR), microsatellite markers or PCR markers. Marker-assisted breeding techniques are useful, in that plants heterozygous for the *bm* allele can be identified without the necessity for evaluating phenotypic ratios in selfed progeny.

Once inbreds having the *bm* phenotype and desired performance characteristics have been identified, each inbred is evaluated for the development of appropriate hybrid combinations by test crosses or top crosses to another inbred displaying the *bm* phenotype.

Suitable hybrids are selected to have certain desirable agronomic characteristics. Such characteristics include, for example, satisfactory disease resistance or tolerance, satisfactory insect resistance or tolerance and satisfactory seedling vigor. Such general characteristics are desired in all types of corn hybrids regardless of the intended use of the hybrid; the best available performance with respect to these characteristics will be incorporated into a *bm* hybrid. Methods and tests for identifying inbreds having the desired general agronomic performance characteristics in hybrid combination are known in the art.

In addition, hybrids are developed that are adapted for use in short, medium or long growing seasons according to a relative maturity rating system such as the Minnesota Maturity Rating (MMR). See, e.g., U.S. Patent 5,495,067, incorporated herein by reference.

However, certain other characteristics are of particular relevance in selecting suitable *bm* inbreds and hybrids. One relevant characteristic is the forage yield of the hybrid. The forage yield of a suitable *bm* hybrid is from about 20 to about 28 Tons per acre (adjusted to 70% moisture), preferably from about 22 Tons per acre to about 28 Tons per acre, more preferably from about 24 Tons per acre to about 28 Tons per acre. The forage yield can be from about 25 Tons/acre to about 32 Tons/acre in newer hybrids that are converted to express the *bm* phenotype.

A *bm* hybrid typically has a decrease in forage yield compared to the forage yield of its isogenic counterpart lacking the *bm* phenotype. However, preferred hybrids have a forage yield decrease of less than 15% compared to their isogenic counterparts, preferably a decrease of about 10% or less.

Another relevant characteristic is the *in vitro* digestibility of corn silage made from a *bm* hybrid as determined after about 30 days of fermentation. *In vitro* true digestibility can be measured by determining neutral detergent fiber (NDF) digestibility. See, e.g., Goering, H. and Van Soest, P., *Forage Fiber analyses in Agriculture Handbook 379*, U.S. Department of Agriculture, Washington, D.C., pp. 1-20 (1975). *In vitro* NDF digestibility is also referred to herein as *in vitro* cell wall digestibility (IVCWD). Neutral detergent fiber is a measure of the cellulose, hemicellulose and lignin fractions of silage and constitutes from about 35 percent to about 55 percent of the silage dry matter, generally from about 40 percent to about 50 percent. Preferred *bm* hybrids have an *in vitro* NDF digestibility of from about 44 percent to about 70 percent, e.g., from about 44 percent to about 60 percent, or from about 47 percent to about 55 percent. In some embodiments, the *in vitro* NDF digestibility is from about 44% to about 50%. The NDF digestibility of *bm* hybrids preferably is about 6 to about 20 percentage units greater than the corresponding isogenic normal hybrid (which is homozygous for *Bm3*), e.g., about 6 to about 15 percentage units greater.

*In vitro* digestibility can also be measured on the whole plant after about 30 days of fermentation. Whole plant *in vitro* digestibility is also referred to herein as IVTD and is based on the Tilley and Terry fermentation method. Tilley, J. and Terry, R. J. *Brit. Grassland Soc. 18:104-111* (1963). Suitable hybrids have an IVTD value of from about 65% to about 85%, e.g., from about 70% to about 85%, or from about 74% to about 85%. In certain embodiments, the IVTD value of *bm* hybrids is from about 74 percent to about 80 percent. The IVTD value of a *bm* hybrid is about 2 to about 7 percentage units greater than the IVTD value of the corresponding isogenic normal hybrid.

*In vitro* NDF digestibility and IVTD can be measured, for example, by collecting fresh-cut plant material and ensiling it in mini-silo fermentation canisters. The material may be reshred prior to ensiling to provide more uniform fermentation. After about 30 days, the pH of the fermented material is stable and *in vitro* NDF Digestibility and IVTD are determined as described in Goering, H. and Van Soest, P., *supra*, except that a 30 hour *in vitro* fermentation is performed rather than a 48 hour *in vitro* fermentation.

The forage yields, IVCWD values and IVTD values of the novel *bm* hybrids disclosed herein are useful in determining the suitability of such hybrids for inclusion in a dairy cattle feed ration.

Once inbreds having the *bm* phenotype and desired performance characteristics have been identified, each inbred

is evaluated for the development of appropriate hybrid combinations by test crosses or top crosses to another inbred displaying the *bm* phenotype.

Examples of corn inbreds suitable for producing *bm* corn hybrids include, without limitation, inbreds *AR5252bm3*, *7675bm3*, *7677bm3*, *AR5251bm3* and *AR5651bm3*. The inbred lines *AR5252bm3*, *AR5251bm3*, *AR5651bm3*, *AR5551bm3*, *AR5153bm3*, *AR5253bm3* and *AR5654bm3* are available from the applicant on request. This however in no way provides a licence of any sort to perform the invention as claimed.

The invention may readily be performed using materials available and known in the art. For example, other corn seeds possessing mutant *bm* alleles are available from various universities and seed stock centers such as Maize Genetics Cooperation - Stock Center, University of Illinois, Department of Crop Science, Urbana, Illinois, USA. These seeds can also be used to initiate a *bm* breeding program.

The invention may alternatively be performed by transfection of appropriate plants to produce antisense molecules to the *Bm* gene whose sequence is known.

An inbred line according to the invention preferably is homozygous for a *bm3* allele. Certain *bm3* alleles have been sequenced, e.g., the *bm3-1* and *bm3-2* alleles. Vignols et al. *Plant Cell* 7:407-416 (1995). Alleles that have a deletion, e.g., a deletion similar to that in *bm3-2*, are preferred because such alleles are less likely to revert to wild-type.

It is known in the art that maize germplasm can be divided into a number of distinct heterotic groupings. Such groups include Reid Yellow Dent, Lancaster Sure Crop and subgroups such as Iowa Stiff Stalk Synthetic (Reid Yellow Dent) and Oh43 (Lancaster Sure Crop). One important aspect of a maize breeding program is the identification of the heterotic group to which a particular inbred belongs. By identifying the heterotic group or subgroup, it becomes possible to more clearly determine the appropriate types of crosses, in order to obtain sufficient levels of heterosis or hybrid vigor. Because of the large number of possible heterotic groupings to which a given inbred can belong, it is useful to ascertain which heterotic groups can be most advantageously used to form a hybrid combination from brown midrib inbreds. Inbreds having the mutant *bm* phenotype have not been tested in all possible heterotic groups for the effect of the *bm* phenotype on expression of the mutant trait and the effect on other agronomic traits.

Once a suitable pair of inbred lines that provide the desired performance in hybrid combination have been identified, production of *F<sub>1</sub>* hybrid *bm* seed is undertaken. Typically, a substantially uniform assemblage of *F<sub>1</sub>* *bm* hybrid corn seeds is conditioned and bagged in packaging material by means known in the art to form an article of manufacture. Alternatively, Such a novel bag of seed has a package label accompanying the bag, e.g., a tag or label secured to the packaging material, a label printed on the packaging material or a label inserted within the bag. The package label indicates that the seeds therein are effective for producing silage that can be fed to dairy cattle. Preferably, the package label indicates that the resulting silage is to be combined with a feed composition component as disclosed herein. The package label may indicate that silage resulting from seeds contained therein is effective for increasing milk production when fed to lactating cows as disclosed herein.

The novel corn seed is planted and cultivated according to standard agronomic practices in the geographic area to which the hybrid is adapted. Growers typically take into account soil fertility, crop rotation practices and other factors specific to the locale in which the hybrid corn is being grown.

Corn plants can be grown to maturity and the seeds produced thereon harvested for use as grain. However, above-ground parts of corn plants disclosed herein preferably are harvested after grain fill, but before drydown. Typically, plants are harvested using a mechanical forage harvester which chops the above-ground portion of the plant into small pieces. Harvesting typically is based on the stage of seed maturity and occurs about the stage at which the color line is halfway down the kernel. The precise harvest time will depend, of course, upon geographical location and seasonal factors such as the weather. The chopped material is then ensiled by techniques known in the art, e.g., in trenches or in concrete stave silos. Microbial inoculants and/or preservatives may be added to promote silage formation, if desired.

Com silage from *bm* plants is fed to ruminant animals, such as dairy cattle as a total animal feed ration, such as a total dairy cattle feed ration (also referred to herein as a total mixed ration or total ration) comprising *bm* silage and a feed composition. The nutrient composition of the silage can be determined, e.g. percent by dry matter, percent NDF, percent CP, *in vitro* NDF, digestibility and IVTD. Based on this information, a total animal (e.g. dairy cattle) feed ration is formulated.

A feed composition component of the invention may be a complete feed form, a concentrate form, blender form or base mix form. By complete feed form it is meant that the feed represents the cow's entire grain ration. By concentrate form it is meant that the feed will be used as the primary supplemental protein source and would normally be fed with grain to meet an animal's protein needs. By blender form it is meant that the feed will be mixed with approximately a 50:50 ratio with grain to form the complete grain ration. The base mix form is similar to the concentrate form, but is typically higher in protein content and is used at lower inclusion rates. The base mix form will be a primary, but often not the sole source of supplemental protein.

For example, a complete feed form composition may contain wheat middlings, corn, meat and bone meal, soybean meal, salt, macro-minerals, trace minerals and vitamins. Alternative or optional ingredients commonly include, but are not restricted to, fat, sunflower meal, feather meal, malt sprouts, distillers' grains, canola meal and soybean hulls. Other

alternative or optional protein sources include, for example, blood meal, corn gluten meal, peanut meal, cottonseed meal, soybeans (extruded or roasted), wheat bran and high fat rice bran.

A concentrate form composition, a blender form composition or a base mix form composition can be prepared by those of skill in the art, based upon the complete feed composition discussed above. Grains fed with the blender, 5 concentrate and base mix forms of the composition can include, but are not limited to, corn, barley, oats, millet, rice, sorghum and wheat. Intake of the total grain ration will typically range from 2.7 to about 17 kilograms (kg) per day.

The nutrient composition of total dairy cattle feed ration comprising *bm* silage contains crude protein (CP) at a 10 relatively high level in order to increase milk production. On a dry matter basis, CP is about 17% or greater, preferably from about 18% to about 21%. If there are reproductive difficulties, the amount of CP may be reduced. About 30% to about 50% of the CP is soluble protein (also referred to as SP, degradable intake protein, or DIP), preferably about 15 35% to about 50%, more preferably about 40% to about 45%. A total ration having a CP level at the lower end of the range of values results in greater weight gain and smaller increases in milk production. On the other hand, a ration with a CP level at the upper end of the range of values results in smaller weight gains and greater increases in milk production. About 20% to about 40% of the CP in the total ration is rumen undegraded protein (RUP, undegradable 20 intake protein, or UIP), preferably from about 25% to about 40%, more preferably from about 25% to about 35%.

The ADF content of a total ration preferably is about 18% or greater, e.g., about 18% to about 24%, or about 19% to 25 about 22%. The fat content of a total ration typically is from about 4% to about 8%, e.g., about 4% to about 6%. The non-fiber carbohydrate (NFC) content of a total ration is typically from about 36% to 46% on a dry matter basis, e.g., about 39% to about 43%.

The forage component in the total ration typically constitutes about 20% to about 60% of the ration on a dry matter 20 basis and is added to achieve a fiber content of about 20% to about 40%, preferably from about 25% to about 35%. These values are appropriate for cows in early lactation; mid-lactation cows may have a higher forage/concentrate 25 ratio resulting in, for example, about 30% to about 40% fiber content. The *bm* silage comprises from about 20% to about 100% of the forage on a dry matter basis, e.g., from about 25% to about 90%, or from about 40% to about 90%, or from about 70% to about 90%. Although *bm* silage may comprise 100% of the forage component in a ration, higher 30 amounts of a protein source such as soybean meal will need to be included; doing so increases the overall cost of the diet. Therefore, it can be more cost-effective to include *bm* silage at less than 100% of the forage component.

Sources that may be used to complete the forage component of the ration include, but are not limited to, corn 35 silage from non-*bm* corn plants, alfalfa haylage, grass silages (e.g., sudangrass, orchardgrass or sorghum-based silage), grass hays (e.g., sudangrass or orchardgrass) and alfalfa or clover hay. Such other forages are known in the art.

The silage component and the feed composition component are combined and fed to ruminant animals, e.g. dairy cattle, under generally accepted management conditions. For example, typical dairy cow management conditions include known measures for animal care, shelter and veterinary treatment, under lactation and gestation cycles used by 35 dairy farmers. Under typical conditions for feeding and managing of lactating dairy cows, a total ration comprising *bm* silage and having a nutrient composition disclosed herein provides a significant increase in milk yield on a raw basis or fat-corrected basis, without adversely affecting general animal health, particularly live weight.

A total ration includes components such as fat, vitamins and minerals in proportions and within ranges that are known in the art. Maximum fiber digestibility occurs in a rumen environment suitable for growth of fiber-digesting organisms. Thus, the levels of other components are considered when formulating a total animal feed ration. For example, 40 high levels of unsaturated fatty acids may depress NDF digestibility. In addition, the level of non-structural carbohydrate is adjusted to provide sufficient energy from starch fermentation without decreasing rumen pH to such an extent that the growth of fiber-digesting microbes is inhibited. Furthermore, the higher digestibility of *bm* corn silage may require that the ration be adjusted to achieve higher levels of non-protein nitrogen (NPN) in the rumen compared to rations using non-*bm* corn silage, because fiber-digesting microbes generally are more active when NPN is increased. Sufficient 45 NPN can be provided by increasing the level and sources of SP in the ration, in combination with an increased forage/concentrate ratio.

Particle size of corn silage affects the rate and the efficiency of silage digestion in the rumen. A theoretical length of cut of about 0.95 cm to 1.27 cm is often used when chopping corn for silage. Because of the higher digestibility of *bm* corn silage, such silage may pass through the rumen more rapidly than desirable for efficient digestion and mat 50 formation. Thus, a larger theoretical length of cut preferably is used when chopping *bm* corn, e.g., about 1.27 cm to about 2.54 cm, or about 1.91 cm to about 2.54 cm. Increased length of cut for *bm* corn silage contributes to increased scratch factor which stimulates cud-chewing and improves rumen buffering due to saliva formation.

From the foregoing it will be appreciated that the relative amounts of different components of the feed may be 55 adjusted appropriately to allow maximum milk production. In particular, where necessary the level of CP should be increased to the upper level of the range given above, the feed should be formulated to maintain rumen pH around 6.2, such as in the range 5.5 to 7.0 for ruminants, particularly 5.5 to 6.5 for dairy cattle, for example by adjustment of the level of non-structural carbohydrate and factors which affect fiber digestibility should be adjusted appropriately, e.g. the levels of unsaturated fatty acids. Modification of each or all of the different components mentioned may be

performed to enhance milk production. Alternatively, or additionally, the particle size of corn silage may be adjusted as mentioned above.

In one embodiment, a method according to the invention comprises administration of a biologically active somatotropin to a ruminant animal such as a dairy cow while feeding a ration comprising *bm* corn silage to the animal. Somatotropin refers to a peptide that has biological activity and chemical structure similar to those of somatotropin produced in the pituitary gland of an animal. Such somatotropin include, without limitation, natural somatotropins produced by pituitary somatotropic cells and somatotropins expressed by genetically transformed microorganisms such as *E. coli* or yeasts. Biologically active somatotropins may have an amino acid sequence identical to natural somatotropins or may be analogs having one or more alterations in amino acid sequence that provide enhanced biological activity or some other advantage. Illustrative embodiments of suitable somatotropins are described in, e.g., US Patents 4,693,973, 5,411,951, 5,037,806 and 5,086,041, all of which are incorporated herein by reference. A preferred somatotropin is bovine somatotropin for use with dairy cows.

Typically, somatotropin is administered parenterally, e.g., by intramuscular or subcutaneous injection, which delivers the peptide to the circulatory system. Suitable formulations for administering a somatotropin are known in the art. For example, somatotropin can be injected in a physiologically acceptable vehicle such as a wax and an oil. See, e.g., U.S. Patents 4,977,140, 5,474,980, 5,156,851 and 5,013,713, all of which are incorporated herein by reference. In some embodiments, a prolonged release formulation is used, such that a dose of somatotropin effective for enhancing milk production over a period of several days is released into the circulatory system with a single administration, e.g., at least 7 days, at least 14 days or at least 21 days.

A biologically active somatotropin is administered under conditions that provide an amount of peptide effective for enhancing milk production to a ruminant animal. Techniques and means are known in the art for determining appropriate doses to be supplied or provided to an individual animal. For example typically, amounts of somatotropin between about 5 and about 75 mg/cow/day are suitable for use in the invention. For example, parenteral administration of a single dose of about 300 mg of zinc-associated somatotropin can be sufficient to provide an increased amount of bovine somatotropin for a period of about 15 days. U.S. Patent 5,086,041, incorporated herein by reference.

A feed ration comprising *bm* corn silage is fed during a selected period to at least one lactating ruminant animal, e.g. dairy cow. Somatotropin is administered so that the production-enhancing effect of the peptide coincides with the feeding of the *bm* silage-containing ration. For example, a prolonged release formulation of somatotropin can be administered to a cow every 14 days over a 180 period. Concomitantly, a *bm* silage ration described above is fed daily to the cow over the same 180 day period. As an alternative, somatotropin can be administered daily and a *bm* silage ration can be fed daily. The somatotropin can be administered, e.g., by injection, either before or during the period in which the ration is being fed, provided that an effective amount of the peptide is delivered during the same period that the ration is being fed. The feeding period can be short, e.g., 4 days, or preferably longer, e.g., 10 days, 30 days or more.

A biologically active somatotropin may be administered to both primiparous and multiparous cows, either in early, mid or late lactation, e.g., about 63 to about 360 days postpartum. Somatotropin is particularly useful for increasing milk production during mid and late lactation; i.e., about 100 to about 360 days postpartum.

Feeding a dairy cattle feed ration comprising *bm* corn silage or *bm* corn silage results in surprising increases in milk production. An increase in milk production of at least about 1.8 kg/cow/day is often observed. A method of feeding a *bm* silage feed ration as disclosed herein provides significant utility to the producer because of such increases in milk production. Moreover, protein, lactose and solids-not-fat (SNF) components of milk are not altered significantly after feeding a *bm* silage total ration as disclosed herein.

Feeding a dairy cattle feed ration comprising *bm* corn silage and coextensively supplying an effective amount of a biologically active somatotropin also results in significant increases in milk production compared to the increase observed with somatotropin alone. For example, an increase in milk production of about 3 kg/cow/day can be observed over a 4 day period.

The invention will be further understood with reference to the following illustrative embodiments, which are purely exemplary and should not be taken as limiting the true scope of the present invention as described in the claims.

#### Example 1

##### **Preparation of Silage from a Brown Midrib Hybrid**

Inbred lines AR5152*bm3* and AR5751*bm3* are crossed (AR5152*bm3* X AR5751*bm3*) to make *F*<sub>1</sub> hybrid seed carrying the brown midrib genotype. Control hybrid seed is prepared by crossing the unconverted parental lines.

The control and *bm* hybrid seed is planted at a density of 24,900 kernels per acre and cultivated using a standard fertilization program. The corn is harvested 4 months after planting, using a forage harvester having a 0.95 cm theoretical length of cut (TLC) and a screen. The harvested material is placed in polyethylene bags (Ag Bags). If desired, the harvested material is treated with inoculant and/or preservative. The forage yield for *bm* corn silage is about 1 wet

ton per acre less than the control hybrid corn.

Samples are collected from the polyethylene bags after 30 days of fermentation/storage and assayed for nutrient concentrations. Typical results are shown in Table 2. The control corn silage and the *bm* corn silage are similar in nutrient composition, except for fiber digestibility and NE<sub>L</sub> concentration. The greater NE<sub>L</sub> concentration is likely related to the *in vitro* digestibility of NDF, ADF, and hemicellulose.

Table 2

Nutrient composition of corn silages <sup>1</sup>			
	Nutrient	Control	BMR
10	Dry matter, %	34.6	31.9
15	Crude protein, %	8.18	8.58
20	Fat, %	2.74	2.59
25	ADF, %	18.76	19.93
30	NDF, %	38.20	40.67
35	Hemicellulose, %	19.44	20.74
	NDF dig., %	52.4	65.3
	ADF dig., %	43.5	60.9
	Hemicellulose dig., %	60.9	69.5
	Ash, %	3.34	3.63
	NFC, %	47.54	44.53
	NE <sub>L</sub> , Mcal/cwt.	76.0	80.5
	Ca, %	.19	.19
	P, %	.23	.22
	Mg, %	.14	.13
	K, %	1.09	1.27
	Na, %	.01	.09
	Salt, %	.22	.13

<sup>1</sup>Nutrients are expressed on a dry matter basis

### Example 2

#### **Formulation of a Dairy Cattle Feed Ration**

The following experimental diets are developed to be fed as a total mixed ration (TMR). The following assumptions are used in ration formulation: the early lactation experimental dairy cow is estimated to weigh 636 kg and to produce 50 kg of milk per day with 3.30% fat, 3.10% milk protein and no body weight change. Dry matter intake (DMI) is predicted to be slightly less than 4.0% of body weight. The TMR is formulated to contain on a dry matter basis: 50% complete feed mix, 33% corn silage and 17% alfalfa haylage. The ingredient composition of the complete feed is given in Table 3 and the nutrient composition of the complete feed mix is given in Table 4.

Table 3

Ingredient Composition of Complete Feed <sup>1</sup>	
INGREDIENT	Composition (% by weight)
Corn, coarse ground	38.7
Wheat Middlings	20.0
Meat & Bone Meal (50% protein)	1.5

<sup>1</sup>As fed basis.

Table 3 (continued)

Ingredient Composition of Complete Feed <sup>1</sup>	
INGREDIENT	Composition (% by weight)
Hi Pro Soybean Meal (49% crude protein)	1.9
Feather Meal	3.5
Canola Meal	20.0
Animal fat	2.3
Minerals and Vitamins	4.7
Distillers grains	7.4
Total	100.0

<sup>1</sup>As fed basis.

Table 4

Nutrient Composition of Complete Feed	
NUTRIENT	COMPOSITION <sup>1</sup>
Dry Matter	88.0
Crude Protein (CP)	23.0
Soluble Protein (SP as % of CP)	21.0
RUP (% of CP)	45.0
Fat	5.0
Net Energy of Lactation (NE <sub>L</sub> ) Mcal/kg	92.0
Acid Detergent Fiber (ADF)	9.3
Neutral Detergent Fiber (NDF)	19.5
Non-Fiber Carbohydrate (NFC)	43.0
Calcium	1.40
Phosphorus	0.90
Magnesium	0.45
Sulfur	0.50
Salt	1.50

<sup>1</sup>Percentages for nutrients given as % by weight on a dry matter basis

45 Brown midrib (BMR) or control corn silage (Example 2) are combined with complete feed mix and alfalfa haylage to form the rations, using each silage in an equal amount on a dry matter basis. Rations contain approximately 18.5% crude protein, 32% soluble protein and 40% rumen undegraded protein. Both rations are adjusted to a calculated pH of about 6.2 Both rations are balanced to meet or exceed mineral, vitamin and total bypass protein requirements.

50 Example 3

**Milk Production Using BMR Silage In a Dairy Cattle Ration**

55 20 lactating dairy cows are utilized in a randomized complete block design experiment, consisting of 5 primiparous cows and 5 multiparous cows fed control silage and 5 primiparous cows and 5 multiparous cows fed BMR silage. Cows are randomly assigned after calving. During the fifth week postpartum, the cows are abruptly switched to the appropriate feeding ration and continued on that ration from week 5 to week 17 postpartum. Primiparous cows are producing more than 25 kg/day of milk during the 4th week postpartum. Multiparous cows are producing greater than 29.5 kg/day of

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5 milk during the 4th week postpartum. All cows are disease free and otherwise healthy. Cows are fed ad libitum for the duration of the experiment, twice daily at 12 hour intervals, if possible. Cows are fed to allow for 5 to 10% feed refusal. The dietary ration and percentages of complete feed mix (Example 2), corn silage (Example 1) and alfalfa haylage on a dry matter basis are given in Table 5.

10 Milk production is measured at each milking and reported daily. Two samples of milk are taken weekly from each cow for composition analysis, including fat, protein, lactose, solids-not-fat and somatic cell counts. Body weights are recorded weekly after calving. Body condition scores are recorded at approximately weeks 4, 8, 12 and 16 postpartum. The same employee records condition scores at each measurement time. Body conditions scores are recorded according to the definitions indicated in Table 6.

15 Table 5

Dietary feeding guidelines to feed concentrate mix and forages					
	Control ration	BMR ration		Control ration	BMR ration
	Ration Content	% of DM	% of DM	DM, %	% as Fed
15	Control Silage	33.0	-	34.6	50.3
20	BMR Silage	-	33.0	31.9	-
	Complete Feed	50.0	50.0	88.0	30.0
	Alfalfa Haylage	17.0	17.0	45.6	19.7
	Total	100.0	100.0		100.0
				100.0	100.0

25 The results indicate that cows fed BMR silage have a statistically significant increase in milk production when comparing unadjusted means or when comparing covariate adjusted means. The increase is from about 1.8 kg milk/cow/day to about 5.0 kg milk/cow/day. However, there is a statistically significant decrease in milkfat percentage in milk from cows fed the ration containing BMR silage.

30 Table 6

Instructions for Body Condition Scoring	
Score	Definition
35 1	1 Loin area has limited flesh covering, is prominent and the ends of spinous processes are sharp to touch. Definite overhanging shelf effect is visible. Individual vertebrae of the hind quarters are prominent and distinct. Hooks and pin bones are notable. The area below the tail-head and between pin bones is severely depressed causing the bone structure of the area to appear extremely sharp.
40 2	2 Individual spinous processes are usually discernible but are not prominent. Ends of processes are sharp to touch, although they have greater flesh covering. The processes do not have a <u>distinct</u> overhanging shelf effect. Individual vertebrae of the hind quarters are not visually distinct but are readily distinguishable by palpitation. Hooks and pin bones are prominent, but the depression between them is less severe. The area below the tail-head and between the pin bones is depressed, but the bone structure is not devoid of flesh covering.
45 3	3 Spinous processes are discernible by applying slight pressure. Area over processes appears smooth and the overhanging shelf effect is not noticeable. Vertebrae of the hindquarters appear as a rounded ridge. Hooks and pin bones are rounded and smooth. The area between the pin bones and around the tail-head appear smooth without sign of fat deposition.
50 4	4 Individual spinous processes can be distinguished only by firm palpitation. Processes appear flat or rounded with no overhanging shelf effect. The ridge formed by the vertebrae of the hindquarters is rounded and smooth, flattening out as you move forward. Hooks are rounded, and the span between hooks is flat. The area around the tail-head and pin bones is rounded with evidence of fat deposition.
55 5	5 Bone structure of the vertebral column, spinous processes, hooks and pin bone regions is not visually apparent. Evidence of fat deposition is prominent. The tail head appears to be buried in fatty tissue.

Example 4**Development of Corn Inbred Lines and Hybrids Having a Brown Midrib Phenotype**

5 Line MED947 is a corn inbred that has the characteristics shown in Table 7. Line MED947 was crossed to B73 $bm3$  (stock no. 980392, obtained from the Department of Agronomy, Purdue University, West Lafayette, Indiana). After completing a conversion program, an inbred line designated as AR5252 $bm3$  was obtained. The backcrossing program is shown in Table 8.

10 Line MED154 is a corn inbred that has the characteristics shown in Table 9. Line MED154 was backcrossed to AR5151 $bm3$  as indicated in Table 10 to obtain line AR5251 $bm3$ . The  $bm3$  allele in AR5151 $bm3$  was obtained from A632 $bm3$  (Purdue University).

Line MED058 is a corn inbred that has the characteristics shown in Table 11. Line MED058 was backcrossed with line AR5654 $bm3$ . The  $bm3$  allele in AR5654 $bm3$  was derived from Mo17 $bm3$  (Purdue University).

15 Line 7675 was crossed to Oh545 $bm3$  (stock no. 980390, Department of Agronomy, Purdue University) in a conversion program as shown in Table 8, resulting in an inbred line designated 7675 $bm3$ .

Line 7677 was crossed to Mo17 $bm3$  (stock no. 980394, Department of Agronomy, Purdue University) in a conversion program as shown in Table 8, resulting in an inbred line designated as 7677 $bm3$ .

Characteristics of the inbred lines AR5252 $bm3$ , 7675 $bm3$  and 7677 $bm3$  are shown in Table 12. Data on flowering for six inbred lines is shown in Table 13.

20 Line AR5252 $bm3$  was crossed as a female to 7675 $bm3$  and to 7677 $bm3$  to form F<sub>1</sub> hybrids 330666 and 330671, respectively. Characteristics of 330666 are shown in Table 14. Characteristics of 330671 are shown in Table 15.

Hybrid checks were prepared from crosses of AR5252 X 7675 and AR5252 X 7677. These F<sub>1</sub> hybrids do not exhibit the brown midrib phenotype.

25 Pairwise comparisons of forage yield, quality profiles and energy values for 330666 and unconverted control hybrid 118629 are shown in Table 16. The same comparisons are shown for 330671 and control hybrid X8311 in Table 17.

Lines AR5751 $bm3$ , AR5654 $bm3$ , AR5253 $bm3$ , AR5153 $bm3$  and AR5551 $bm3$  were developed by backcrossing programs similar to those described above. Each of these lines exhibits the brown midrib phenotype. Characteristics of these lines are shown in Tables 18-23.

30

Table 7

Description of Line MED947				
COTYLEDON LEAF		EAR CHARACTERISTICS		
35 ANTHOCYANIN LENGTH	N/A	SILK COLOR	YELLOW	
	N/A	EAR LEAVES	ABSENT	
PLANT AND STALK		HUSK		
40 PLANT HEIGHT	7.50	COVERAGE	COVERS TIP	
EAR HEIGHT	3.00	LOOSENESS	TIGHT	
UNIFORMITY RATING	8	EAR ANGLE RATING	6	
ANTHOCY. IN BRACE ROOTS	PRESENT	SHANK LENGTH	<4in.	
ANTHOCY. IN NODES	ABSENT	EAR LENGTH	6-9in.	
DIAM. AT 2nd NODE	0.00	EAR SHAPE	CYLINDRICAL	
45 ROOT RATING	N/A	UNIFORMITY	8	
LEAVES		KERNEL		
ANGLE	UPRIGHT	NO. ROWS	16	
50 COLOR	MEDIUM GREEN	TYPE	SEMI DENT	
TOTAL NUMBER		SIZE	AVERAGE	
NUMBER ABOVE EAR	5-6	BODY COLOR	YELLOW	
LENGTH, EAR LEAF	0	CROWN COLOR	LIGHT YELLOW	
MAX., WIDTH, EAR LEAF	0.00			
55 ANTHOCYANIN, MARGIN	ABSENT	COB		
ANTHOCYANIN, SHEATH	ABSENT	COLOR		
PUBESCEENCE, SHEATH	PRESENT		PINK	

Table 7 (continued)

Description of Line MED947			
<u>COTYLEDON LEAF</u>		<u>EAR CHARACTERISTICS</u>	
5	PUBESCEENCE, MARGINS PLANT, TILLERING TASSEL	PRESENT 0.00	DIAMETER MIDPOINT < 1 in.
10	<u>TASSEL</u> COMPACTNESS BRANCH ANGLE NUMBER PRIMARY BRANCHES SECONDARY BRANCH	COMPACT >60 4-8 PRESENT	INSECT RESISTANCE ATTRACTIVE TO APHIDS N/A
15	LENGTH SIZE RATING	N/A 7	DISEASE RESISTANCE RATINGS NORTHERN LEAF BLIGHT(R2) SOUTHERN LEAF BLIGHT(R0) NORTHERN COB LEAF SPOT (R3)
20	TASSEL EXTENSION PARTIALLY SHED IN BOOT? DIFFICULTY IN PULLING NUMBER LEAVES PULLED	ENCLOSED NO AVERAGE 0	STEWARTS BACTERIAL WILT GREY LEAF SPOT EYESPOT ANTHRACNOSE
25	ANTHER COLOR GLUME COLOR - TIP - BASE - BAND POLEN SHED DURATION POLEN AMOUNT RATING	YELLOW RED LIGHT GREEN RED AVERAGE AVERAGE	(9=Resistant, 1=Susceptible) (Letter indicates confidence level)

30

Table 8  
Inbred Line Conversion to Brown MidRibAR5252bm3

35	(MED947XB73bm <sub>3</sub> )MED947BC <sub>6</sub> (MED947XB73bm <sub>3</sub> )MED947BC <sub>5</sub> S <sub>1</sub> (MED947XB73bm <sub>3</sub> )MED947BC <sub>5</sub> (MED947XB73bm <sub>3</sub> )MED947BC <sub>4</sub> (MED947XB73bm <sub>3</sub> )MED947BC <sub>3</sub> S <sub>1</sub> (MED947XB73bm <sub>3</sub> )MED947BC <sub>3</sub>
40	(MED947XB73bm <sub>3</sub> )MED947BC <sub>2</sub> (MED947XB73bm <sub>3</sub> )MED947BC <sub>1</sub> S <sub>1</sub> (MED947XB73bm <sub>3</sub> )MED947BC <sub>1</sub> (MED947XB73bm <sub>3</sub> )F <sub>1</sub> MED947XB73bm <sub>3</sub> )F <sub>0</sub>
45	

7675bm3

50	(7675xOh545bm <sub>3</sub> )7675BC <sub>6</sub> S <sub>1</sub> (7675xOh545bm <sub>3</sub> )7675BC <sub>6</sub> (7675xOh545bm <sub>3</sub> )7675BC <sub>5</sub> (7675xOh545bm <sub>3</sub> )7675BC <sub>4</sub> (7675xOh545bm <sub>3</sub> )7675BC <sub>3</sub> S <sub>1</sub> (7675xOh545bm <sub>3</sub> )7675BC <sub>3</sub> (7675xOh545bm <sub>3</sub> )7675BC <sub>2</sub> (7675xOh545bm <sub>3</sub> )7675BC <sub>1</sub> S <sub>1</sub> (7675xOh545bm <sub>3</sub> )7675BC <sub>1</sub>
55	

Table 8 (continued)  
Inbred Line Conversion to Brown MidRib

7675bm3

5 (7675xOh545bm<sub>3</sub>)7675F<sub>1</sub>  
(7675xOh545bm<sub>3</sub>)F<sub>0</sub>

7677bm3

10 (7677XMo17bm<sub>3</sub>)7677BC<sub>5</sub>S<sub>1</sub>  
(7677XMo17bm<sub>3</sub>)7677BC<sub>5</sub>  
(7677XMo17bm<sub>3</sub>)7677BC<sub>4</sub>  
(7677XMo17bm<sub>3</sub>)7677BC<sub>3</sub>S<sub>1</sub>  
15 (7677XMo17bm<sub>3</sub>)7677BC<sub>3</sub>  
(7677XMo17bm<sub>3</sub>)7677BC<sub>2</sub>  
(7677XMo17bm<sub>3</sub>)7677BC<sub>1</sub>S<sub>1</sub>  
(7677XMo17bm<sub>3</sub>)7677BC<sub>1</sub>  
20 (7677XMo17bm<sub>3</sub>)F<sub>1</sub>  
(7677XMo17bm<sub>3</sub>)F<sub>0</sub>

Table 9

Description of Line MED154				
<u>COTYLEDON LEAF</u>		<u>EAR CHARACTERISTICS</u>		
ANTHOCYANIN LENGTH	N/A <2	SILK COLOR EAR LEAVES <u>HUSK</u>	RED MEDIUM	
<u>PLANT AND STALK</u>		COVERAGE LOOSENESS	COVERS TIP LOOSE	
PLANT HEIGHT	7.0			
EAR HEIGHT	2.0			
UNIFORMITY RATING	7	EAR ANGLE RATING	6	
ANTHOCY. IN BRACE ROOTS	PRESENT	SHANK LENGTH	4-12	
ANTHOCY. IN NODES	ABSENT	EAR LENGTH	6-9	
DIAM. AT 2nd NODE	1.0	EAR SHAPE	CONICAL	
ROOT RATING	AVERAGE	UNIFORMITY	7	
<u>LEAVES</u>		<u>KERNEL</u>		
ANGLE	VERY	NO. ROWS	16	
COLOR	MEDIUM	TYPE	DENT	
TOTAL NUMBER	10-15	SIZE	AVERAGE	
NUMBER ABOVE EAR	5-6	BODY COLOR	ORANGE	
LENGTH, EAR LEAF	34	CROWN COLOR	YELLOW	
MAX., WIDTH, EAR LEAF	4.50			
ANTHOCYANIN, MARGIN	PRESENT	<u>COB</u>		
ANTHOCYANIN, SHEATH	ABSENT	COLOR	WHITE	
PUBESCENCE, SHEATH	ABSENT	DIAMETER	1-2	
PUBESCENCE, MARGINS	PRESENT	MIDPOINT		
PLANT, TILLERING TASSEL	0.00			
<u>TASSEL</u>		<u>INSECT RESISTANCE</u>		
COMPACTNESS	AVERAGE	ATTRACTIVE TO APHIDS	YES	
BRANCH ANGLE	30-60			

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Table 9 (continued)

Description of Line MED154			
5	NUMBER PRIMARY BRANCHES	<4	
	SECONDARY BRANCH	ABSENT	
	LENGTH	10-14	
	SIZE RATING	2	
10	TASSEL EXTENSION	PARTIALLY ENCLOSED	
	TASSEL FERTILITY	0	
	SHED IN BOOT?	NO	
	DIFFICULTY IN PULLING	AVERAGE	
	NUMBER LEAVES PULLED	1	
15	ANTHER COLOR	PINK	
	GLUME COLOR - TIP	LIGHT GREEN	
	- BASE	RED	
	- BAND	DARK GREEN	
	POLLEN SHED DURATION	SHORT	
20	POLLEN AMOUNT RATING	LONG	

Table 10

Inbred Line Conversion to Brown Midrib

AR5251bm3

25 (MED154XAR5151bm<sub>3</sub>)C<sub>5</sub>  
 (MED154XAR5151bm<sub>3</sub>)C<sub>4</sub>  
 (MED154XAR5151bm<sub>3</sub>)C<sub>3</sub>S<sub>1</sub>  
 30 (MED154XAR5151bm<sub>3</sub>)C<sub>3</sub>  
 (MED154XAR5151bm<sub>3</sub>)C<sub>2</sub>  
 (MED154XAR5151bm<sub>3</sub>)C<sub>1</sub>S<sub>1</sub>  
 (MED154XAR5151bm<sub>3</sub>)C<sub>1</sub>  
 35 (MED154XAR5151bm<sub>3</sub>)F<sub>1</sub>  
 (MED154XAR5151bm<sub>3</sub>)F<sub>0</sub>

AR5651bm3

40 (MED058XAR5654bm<sub>3</sub>)C<sub>5</sub>S<sub>1</sub>  
 (MED058XAR5654bm<sub>3</sub>)C<sub>5</sub>  
 (MED058XAR5654bm<sub>3</sub>)C<sub>4</sub>  
 (MED058XAR5654bm<sub>3</sub>)C<sub>3</sub>  
 45 (MED058XAR5654bm<sub>3</sub>)C<sub>2</sub>  
 (MED058XAR5654bm<sub>3</sub>)C<sub>1</sub>S<sub>1</sub>  
 (MED058XAR5654bm<sub>3</sub>)C<sub>1</sub>  
 (MED058XAR5654bm<sub>3</sub>)F<sub>1</sub>  
 (MED058XAR5654bm<sub>3</sub>)F<sub>0</sub>

50

Table 11

Description of Line MED058			
COTYLEDON LEAF		EAR CHARACTERISTICS	
55 ANTHOCYANIN	N/A	SILK COLOR	YELLOW
LENGTH	<2	EAR LEAVES	ABSENT

Table 11 (continued)

Description of Line MED058				
5	<u>PLANT AND STALK</u>		<u>HUSK</u>	
	PLANT HEIGHT	6.5	COVERAGE	
	EAR HEIGHT	3.0	LOOSENESS	AVERAGE
	UNIFORMITY RATING	7	EAR ANGLE RATING	5
10	ANTHOCY. IN BRACE ROOTS	PRESENT	SHANK LENGTH	<4
	ANTHOCY. IN NODES	ABSENT	EAR LENGTH	6-9
	DIAM. AT 2nd NODE	1.0	EAR SHAPE	
	ROOT RATING	AVERAGE	UNIFORMITY	CYLINDRICAL
				9
15	<u>LEAVES</u>		<u>KERNEL</u>	
	ANGLE	UPRIGHT	NO. ROWS	12
	COLOR	LIGHT GREEN	TYPE	DENT
	TOTAL NUMBER	10-15	SIZE	AVERAGE
20	NUMBER ABOVE EAR	>6	BODY COLOR	ORANGE
	LENGTH, EAR LEAF	21	CROWN COLOR	YELLOW
	MAX., WIDTH, EAR LEAF	3.00		
	ANTHOCYANIN, MARGIN	PRESENT		
	ANTHOCYANIN, SHEATH	ABSENT	<u>COB</u>	
25	PUBESCENCE, SHEATH	ABSENT	COLOR	WHITE
	PUBESCENCE, MARGINS	ABSENT	DIAMETER	<1
	PLANT, TILLERING TASSEL	0.00	MIDPOINT	
30	<u>TASSEL</u>		<u>INSECT RESISTANCE</u>	
	COMPACTNESS	LOOSE	ATTRACTIVE TO APHIDS	NO
	BRANCH ANGLE	>60		
	NUMBER PRIMARY BRANCHES	4-6		
35	SECONDARY BRANCH	ABSENT		
	LENGTH	>14		
	SIZE RATING	5		
	TASSEL EXTENSION	PARTIALLY ENCLOSED		
	TASSEL FERTILITY	0		
40	SHED IN BOOT?	NO		
	DIFFICULTY IN PULLING	EASY		
	NUMBER LEAVES PULLED	2		
	ANTHER COLOR	YELLOW		
45	GLUME COLOR - TIP	LIGHT GREEN		
	- BASE	LIGHT GREEN		
	- BAND	DARK GREEN		
	POLLEN SHED DURATION	AVERAGE		
	POLLEN AMOUNT RATING	LIGHT		

50

55

Table 12  
Description of Line 7675bm3

7675bm3

<u>COTYLEDON LEAF</u>		<u>EAR CHARACTERISTICS</u>		
ANTHOCYANIN	PRESENT	SILK COLOR	YELLOW	
LENGTH	2-3	EAR LEAVES	ABSENT	
		HUSK		
<u>PLANT AND STALK</u>		COVERAGE	SHORT	
PLANT HEIGHT	5.5	LOOSENESS	N/A	
EAR HEIGHT	3.0			
UNIFORMITY RATING	2	EAR ANGLE RATING	8	
ANTHOCY. IN BRACE ROOTS	PRESENT	SHANK LENGTH	4-12	
ANTHOCY. IN NODES	ABSENT	EAR LENGTH	6-9	
DIAM. AT 2nd NODE	1.1	EAR SHAPE	CYLINDRICAL	
ROOT RATING	EXCELLENT	UNIFORMITY	7	
<u>LEAVES</u>		<u>KERNEL</u>		
ANGLE	UPRIGHT	NO. ROWS	8-10	
COLOR	VARIEGATED	TYPE	SEMI DENT	
TOTAL NUMBER	10-15	SIZE	AVERAGE	
NUMBER ABOVE EAR	5-6	BODY COLOR	ORANGE	
LENGTH, EAR LEAF	26	CROWN COLOR	YELLOW	
MAX., WIDTH, EAR LEAF	3.50			
ANTHOCYANIN, MARGIN	ABSENT	<u>COB</u>		
ANTHOCYANIN, SHEATH	ABSENT	COLOR	WHITE	
PUBESCENCE, SHEATH	ABSENT	DIAMETER MIDPOINT	<1	
PUBESCENCE, MARGINS	ABSENT			
PLANT, TILLERING TASSEL	0.00			
<u>TASSEL</u>		<u>INSECT RESISTANCE</u>		
COMPACTNESS	AVERAGE	ATTRACTIVE TO APHIDS	NO	
BRANCH ANGLE	30-60			
NUMBER PRIMARY BRANCHES	4-8			
SECONDARY BRANCH	ABSENT			
LENGTH	10-14			
TASSEL FERTILITY	1			
SIZE RATING	6			
TASSEL EXTENSION	PARTIALLY ENCLOSED			
SHED IN BOOT?	NO			
DIFFICULTY IN PULLING	AVERAGE			
NUMBER LEAVES PULLED	1			
ANTHER COLOR	YELLOW			
GLUME COLOR - TIP	LIGHT GREEN			
- BASE	YELLOW			
- BAND	DARK GREEN			
POLLEN SHED DURATION	SHORT			
POLLEN AMOUNT RATING	HEAVY			

Table 12 Cont'd

7677bm3

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<u>COTYLEDON LEAF</u>		<u>EAR CHARACTERISTICS</u>		
ANTHOCYANIN	PRESENT	SILK COLOR	YELLOW	
LENGTH	2-3	EAR LEAVES	ABSENT	
		HUSK		
<u>PLANT AND STALK</u>		COVERAGE	LONG	
PLANT HEIGHT	5.5	LOOSENESS	N/A	
EAR HEIGHT	1.0			
UNIFORMITY RATING	8	EAR ANGLE RATING	6	
ANTHOCY. IN BRACE ROOTS	PRESENT	SHANK LENGTH	<4	
ANTHOCY. IN NODES	ABSENT	EAR LENGTH	6-9	
DIAM. AT 2nd NODE	1.1	EAR SHAPE	CYLINDRICAL	
ROOT RATING	EXCELLENT	UNIFORMITY	7	
<u>LEAVES</u>		<u>KERNEL</u>		
ANGLE	DROOPING	NO. ROWS	12	
COLOR	LIGHT GREEN	TYPE	SEMI DENT	
TOTAL NUMBER	10-15	SIZE	AVERAGE	
NUMBER ABOVE EAR	5-6	BODY COLOR	ORANGE	
LENGTH, EAR LEAF	31	CROWN COLOR	YELLOW	
MAX., WIDTH, EAR LEAF	3.00			
ANTHOCYANIN, MARGIN	ABSENT	<u>COB</u>		
ANTHOCYANIN, SHEATH	ABSENT	COLOR	PINK	
PUBESCENCE, SHEATH	ABSENT			
PUBESCENCE, MARGINS	PRESENT	DIAMETER MIDPOINT	1-2	
PLANT, TILLERING TASSEL	0.00			
<u>TASSEL</u>		<u>INSECT RESISTANCE</u>		
COMPACTNESS	AVERAGE	ATTRACTIVE TO APHIDS	NO	
BRANCH ANGLE	30-60			
NUMBER PRIMARY BRANCHES	4-8			
SECONDARY BRANCH	ABSENT			
LENGTH	>14			
TASSEL FERTILITY	5			
SIZE RATING	7			
TASSEL EXTENSION	PARTIALLY ENCLOSED			
SHED IN BOOT?	NO			
DIFFICULTY IN PULLING	EASY			
NUMBER LEAVES PULLED	1			
ANTHER COLOR	YELLOW			
GLUME COLOR - TIP	RED			
- BASE	RED			
- BAND	DARK GREEN			
POLLEN SHED DURATION	N/A			
POLLEN AMOUNT RATING	LONG			

Table 12 Cont'd

AR5252bm3

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<u>COTYLEDON LEAF</u>		<u>EAR CHARACTERISTICS</u>		
ANTHOCYANIN LENGTH	PRESENT 2-3	SILK COLOR	YELLOW	
		EAR LEAVES	ABSENT	
		HUSK		
PLANT AND STALK		COVERAGE	COVERS TIP	
PLANT HEIGHT	6.0	LOOSENESS	N/A	
EAR HEIGHT	2.0			
UNIFORMITY RATING	4	EAR ANGLE RATING	6	
ANTHOCY. IN BRACE ROOTS	PRESENT	SHANK LENGTH	4-12	
ANTHOCY. IN NODES	ABSENT	EAR LENGTH	6-9	
DIAM. AT 2nd NODE	1.1	EAR SHAPE	CYLINDRICAL	
ROOT RATING	EXCELLENT	UNIFORMITY		8
LEAVES		<u>KERNEL</u>		
ANGLE	UPRIGHT	NO. ROWS	16	
COLOR	MEDIUM	TYPE	DENT	
TOTAL NUMBER	10-15	SIZE	AVERAGE	
NUMBER ABOVE EAR	5-6	BODY COLOR	ORANGE	
LENGTH, EAR LEAF	30	CROWN COLOR	YELLOW	
MAX., WIDTH, EAR LEAF	4.00			
ANTHOCYANIN, MARGIN	ABSENT	<u>COB</u>		
ANTHOCYANIN, SHEATH	PRESENT	COLOR	PINK	
PUBESCENCE, SHEATH	ABSENT	DIAMETER MIDPOINT	1-2	
PUBESCENCE, MARGINS	ABSENT			
PLANT, TILLERING TASSEL	0.00			
TASSEL		<u>INSECT RESISTANCE</u>		
COMPACTNESS	COMPACT	ATTRACTIVE TO APHIDS	NO	
BRANCH ANGLE	<30			
NUMBER PRIMARY BRANCHES	4-8			
SECONDARY BRANCH	PRESENT			
LENGTH	>14			
TASSEL FERTILITY	7			
SIZE RATING	4			
TASSEL EXTENSION	PARTIALLY ENCLOSED			
SHED IN BOOT?	NO			
DIFFICULTY IN PULLING	EASY			
NUMBER LEAVES PULLED	1			
ANTHER COLOR	YELLOW			
GLUME COLOR - TIP	RED			
- BASE	YELLOW			
- BAND	DARK GREEN			
POLLEN SHED DURATION	SHORT			
POLLEN AMOUNT RATING	LONG			

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## Parental Description Legend

## COTYLEDON LEAF

5      Anthocyanin (Present, Absent)  
       Length (<2, 2-3, >3 inches)

10     PLANT AND STALK  
       Plant Height (in feet)  
       Ear Height (in feet)  
       Uniformity (9:most uniform; 1:least uniform)  
       Anthocyanin In Brace Roots (Absent,Present,Dark)  
       Anthocyanin In Nodes (Ab,Present)  
       Stk Diameter at 2nd Node (in inches)  
       Root Rating (9=Good Roots, 5=Average Roots, 1=Bad Roots)

15     LEAVES  
       Angle (Very Erect, Upright, Horizontal, Drooping)  
       Color (Dark Green, Medium Green, Light Green, Variegated)  
       Total # (<9, 10-15, >15)  
       # Above Ear (<5, 5-6, >6)  
       Length, Ear Leaf (in inches)  
       Max. Width, Ear Leaf (in inches)  
       Anthocyanin, Margin (Absent,Present)  
       Anthrocyanin, Sheath (Absent,Present)  
       Pubescence, Sheath (Absent,Present,Very Hairy)  
       Pubescence, Margins (Absent,Present)  
       Plants, Tillering Tassel (%) Percentage

## TASSEL

30     Compactness (Loose,Average,Compact)  
       Branch Angle (<30, 30-60, >60)  
       #, Primary Branches (<4, 4-8, >8)  
       Secondary Branches (Absent,Present)  
       Length (<10 in., 10-14, >14 in.)  
       Size (9:largest; 1:smallest)  
       Shed in Boot (Yes, No)  
       Tassel Extension (Enclosed, Partially Enclosed, Open)  
       Tassel Fertility (9=Male Sterile, 7=Male Sterile, Anthers  
                           Extruded and No Pollen, 5=Intermediate  
                           With Some Viable and Nonviable Pollen,  
                           3=Near Normal Pollen, 1=Completely Normal  
                           Pollen Shed and Viability)

35     Difficulty in Pulling (Easy, Average, Hard)  
       # Leaves Pulled (Ave)  
       Anther Color (Yellow, Green, Pink, Red, Purple)

40     Glume Color  
       Tip (Green, Light Green, Red, Yellow)  
       Base (Green, Light Green, Red, Yellow)  
       Banded (Dark Green, Red, Purple)

45     Pollen shed Duration (Short, Average, Long)  
       Pollen Amount (Low, Average, Heavy)

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## Parental Description Legend Cont'd

## EAR CHARACTERISTICS

5                   Silk Color (Green, Yellow, Pink, Red, Purple)  
                   Ear Leaves (Absent, Medium, Long)  
                   Husk  
                     Coverage (Short, Covers tip, Long)  
                     Looseness (Loose, Average, Tight)  
 10                 Ear Angle at Harvest (9:sharpest; 1:lowest)  
                   Shank Length (<4, 4-12, >12)  
                   Ear Length (<6, 6-9, >9)  
                   Ear Shape (Cylindrical, Conical, Cylindrical/Conical)  
                   Uniformity Rating (9:most uniform; 1:least uniform)  
 15                 Kernel  
                     No. Rows (8-10, 12, 14, 16, >18)  
                     Type (Flint, Semi dent, Dent, Rough dent)  
                     Size (Small, Average, Large)  
 20                 Body Color (White, Light Yellow, Yellow, Orange, Red)  
                     Crown Color (White, Light Yellow, Yellow, Orange, Red)  
                   Cob  
                     Color (Red, Pink, White)  
                     Diameter Midpoint (<1, 1-2, >2)  
 25                 INSECT RESISTANCE  
                     Attractive to Aphids (Yes, Average, No)

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Table 13

Flowering Characteristics of <i>bm</i> Inbred Lines					
	7675 <i>bm3</i>		AR5252 <i>bm3</i>		7677 <i>bm3</i>
35	Slk Heat 1	1,564	Slk Heat 1	1,534	Slk Heat 1
	Slk Heat M	1,587	Slk Heat M	1,607	Slk Heat M
	Slk Heat F	1,633	Slk Heat F	1,587	Slk Heat F
	Slk Days 1	86	Slk Days 1	85	Slk Days 1
	Slk Days M	87	Slk Days M	88	Slk Days M
	Slk Days F	89	Slk Days F	87	Slk Days F
40	Pol Heat 1	1,534	Pol Heat 1	1,505	Pol Heat 1
	Pol Heat M	1,587	Pol Heat M	1,534	Pol Heat M
	Pol Heat F	1,607	Pol Heat F	1,587	Pol Heat F
	Pol Days 1	85	Pol Days 1	84	Pol Days 1
	Pol Days M	87	Pol Days M	85	Pol Days M
	Pol Days F	88	Pol Days F	87	Pol Days F
45	AR5651 <i>bm3</i>			AR5251 <i>bm3</i>	
	Slk Heat 1	1,387	Slk Heat 1	1,552	
	Slk Heat M	1,437	Slk Heat M	1,582	
	Slk Heat F	1,479	Slk Heat F	1,656	
	Slk Days 1	83	Slk Days 1	91	
	Slk Days M	85	Slk Days M	93	
50	Slk Days F	87	Slk Days F	97	
	AR5651 <i>bm3</i>			AR5251 <i>bm3</i>	
	Slk Heat 1	1,387	Slk Heat 1	1,552	
	Slk Heat M	1,437	Slk Heat M	1,582	
	Slk Heat F	1,479	Slk Heat F	1,656	
	Slk Days 1	83	Slk Days 1	91	
55	Slk Days M	85	Slk Days M	93	
	Slk Days F	87	Slk Days F	97	

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Table 13 (continued)

Flowering Characteristics of <i>bm</i> Inbred Lines			
AR5651 <i>bm3</i>		AR5251 <i>bm3</i>	
Pol Heat 1	1,304	Pol Heat 1	1,539
Pol Heat M	1,387	Pol Heat M	1,552
Pol Heat F	1,459	Pol Heat F	1,596
Pol Days 1	79	Pol Days 1	90
Pol Days M	83	Pol Days M	91
Pol Days F	86	Pol Days F	94

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Trait Summary for 330666			
	RATING	SCALE	DEFINITION
Trait:			
Yield for Maturity (9-1)	9	9-1	9=GOOD 1=POOR
Stalk Strength (9-1)	Acceptable	9-1	9=GOOD 1=POOR
Root Strength-Summer (9-1)	9	9-1	9=GOOD 1=POOR
Root Strength-Fall (9-1)	NA	9-1	9=GOOD 1=POOR
Testweight (9-1)	NA	9-1	9=GOOD 1=POOR
Drydown (9-1)	NA	9-1	9=GOOD 1=POOR
Ear Retention (9-1)	9	9-1	9=GOOD 1=POOR
Early Vigor (9-1)	5	9-1	9=GOOD 1=POOR
Seedling Purple Color (9-1)	4	9-1	9=NO PURPLE 1=DARK PURPLE
Greensnap Potential (9-1)	9	9-1	9=LOW 1=HIGH
Drought Stress (9-1)	7	9-1	9=GOOD 1=POOR
Stay Green (9-1)	8	9-1	9=GOOD 1=POOR
Plant Health (9-1)	7	9-1	9=GOOD 1=POOR
Recommended Population (H M L)	H	H,M,L	HIGH, MEDIUM, LOW
Plant Height (S MS M MT T)	8.0	S,MS,M,MT,T	SHORT/MSHORT/MODERATE/MTALL/TA
Ear Height (S MS M MT T)	3.5	S,MS,M,MT,T	SHORT/MSHORT/MODERATE/MTALL/TA
Cargill RM (# Days)	115 (NA)	DAYs	# DAYS
Flowering Date (E M L)	M	E,M,L	EARLY, MEDIUM, LATE
GDU's to Mid-Silk (#)	1464	#	# UNITS
GDU's to Black Layer (#)	2594	#	# UNITS

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Table 14 (continued)

Trait Summary for 330666				
	RATING	SCALE	DEFINITION	
5	Trait:			
Plant Color (L M D)	M	L,M,D	LIGHT,MEDIUM,DARK	
Leaf Angle (E U H D)	U	E,U,H,D	ERECT/UPRIGHT/HORIZONTAL.DROOPY	
10 Leaf Width (W M N)	M	W,M,N	WIDE,MEDIUM,NARROW	
Tillers (9-1)	8	9-1	9=NONE 1=MANY	
Shank Length (9-1)	5	9-1	9=SHORT 1=LONG	
15 Husk Coverage (9-1)	8	9-1	9=GOOD 1=POOR	
Ear Tip Fill (9-1)	6	9-1	9=GOOD 1=POOR	
Ear Length (L M S)	M	L,M,S	LONG,MEDIUM,SHORT	
20 Ear Girth (G M S)	M	G,M,S	GIRTHY,MEDIUM,SLENDER	
Ear Flex (9-1)	NA	9-1	9=GOOD 1=POOR	
Kernel Rows (# Range)	14-16	#	# Range	
Kernel Depth (S M D)	D	S,M,D	SHALLOW,MODERATE,DEEP	
25 Cob Color (R P W)	P	R,P,W	RED,PINK,WHITE	
Kernel Color (W Y G B O R)	Y	W,Y,G,B,O,R	WHITE,YLLW,GLD,BRNZ,ORNG,RED	
Kernel Texture (F SF SD D)	SD	F,SF,SD,D	FLINT,SEMI FLINT,SEMIDENT,DENT	
30				

Table 15

Trait Summary for 330671				
	RATING	SCALE	DEFINITION	
35	Trait:			
Yield for Maturity (9-1)	7	9-1	9=GOOD 1=POOR	
40 Stalk Strength (9-1)	Acceptable	9-1	9=GOOD 1=POOR	
Root Strength-Summer (9-1)	9	9-1	9=GOOD 1=POOR	
Root Strength-Fall (9-1)	NA	9-1	9=GOOD 1=POOR	
45 Testweight (9-1)	NA	9-1	9=GOOD 1=POOR	
Drydown (9-1)	NA	9-1	9=GOOD 1=POOR	
Ear Retention (9-1)	9	9-1	9=GOOD 1=POOR	
50 Early Vigor (9-1)	5	9-1	9=GOOD 1=POOR	
Seedling Purple Color (9-1)		9-1	9=NO PURPLE 1=DARK PURPLE	
Greensnap Potential (9-1)	8	9-1	9=LOW 1=HIGH	
Drought Stress (9-1)	8	9-1	9=GOOD 1=POOR	
55 Stay Green (9-1)	8	9-1	9=GOOD 1=POOR	
Plant Health (9-1)	8	9-1	9=GOOD 1=POOR	
Recommended Population (H M L)	H	H,M,L	HIGH, MEDIUM, LOW	
Plant Height (S MS M MT T)	8.5	S,MS,M,MT,T	SHORT/MSHORT/MODERATE/MTALL/TA	

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Table 15 (continued)

Trait Summary for 330671				
	RATING	SCALE	DEFINITION	
5	Trait:			
	Ear Height (S MS M MT T)	3.0	S,MS,M,MT,T	SHORT/MSHORT/MODERATE/MTALL/TA
10				
	Cargill RM (# Days)		DAYs	# DAYS
	Flowering Date (E M L)	M	E,M,L	EARLY, MEDIUM, LATE
15	GDU's to Mid-Silk (#)	1529	#	# UNITS
	GDU's to Black Layer (#)	NA	#	# UNITS
20	Plant Color (L M D)		L,M,D	LIGHT, MEDIUM, DARK
	Leaf Angle (E U H D)		E,U,H,D	ERECT/UPRIGHT/HORIZONTAL/DROOPY
	Leaf Width (W M N)		W,M,N	WIDE, MEDIUM, NARROW
25	Tillers (9-1)		9-1	9=NONE 1=MANY
	Shank Length (9-1)		9-1	9=SHORT 1=LONG
	Husk Coverage (9-1)		9-1	9=GOOD 1=POOR
	Ear Tip Fill (9-1)		9-1	9=GOOD 1=POOR
30	Ear Length (L M S)		L,M,S	LONG, MEDIUM, SHORT
	Ear Girth (G M S)		G,M,S	GIRTHY, MEDIUM, SLENDER
	Ear Flex (9-1)	NA	9-1	9=GOOD 1=POOR
35	Kernel Rows (# Range)		#	# Range
	Kernel Depth (S M D)		S,M,D	SHALLOW, MODERATE, DEEP
	Cob Color (R P W)	P	R,P,W	RED, PINK, WHITE
	Kernel Color (W Y G B O R)		W,Y,G,B,O,R	WHITE, YLLW, GLD, BRNZ, ORNG, RED
40	Kernel Texture (F SF SD D)		F,SF,SD,D	FLINT, SEMIFLINT, SEMIDENT, DENT

Table 16

Pairwise Comparison of 330666 and 118629 <sup>a</sup>				
Traits	330666	118629	Range	#Loc.
<b>Plant Features</b>				
Plant Hgt	45.54	47.18	-1.64	5
Ear Hgt	18.80	19.80	-1.00	5
<b>Yield Profile<sup>b</sup></b>				
70% Tons/A	25.00	27.49	-2.49	6
DM Tons/A	7.50	8.25	-0.75	6

<sup>a</sup>Summary over two years

<sup>b</sup>DM=Dry Matter; DDM=Digestible Dry Matter; Tons/A=Tons per acre

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Table 16 (continued)

Pairwise Comparison of 330666 and 118629 <sup>a</sup>					
	Traits	330666	118629	Range	#Loc.
<b>Yield Profile<sup>b</sup></b>					
	DDM Tons/A	5.90	6.07	-0.17	6
<b>Quality Profile<sup>c</sup></b>					
	% ADF	23.18	24.83	-1.65	6
	% NDF	43.56	45.08	-1.52	6
	% ASH	4.62	4.51	0.11	6
	% CP	8.92	8.29	0.63	6
	% IVTD	78.75	73.26	5.49	6
	% IVCWD	50.99	40.64	10.35	6
	% WP Mst	70.83	66.54	4.29	6
	% WP DM	29.17	33.46	-4.29	6
<b>Energy Values<sup>d</sup></b>					
	TDN	67.97	66.28	1.69	6
	NEL (Mcal/LB)	0.69	0.67	0.02	6
	NFC%	39.80	39.02	0.78	6
	Lbs CP/A	1,330.26	1,355.60	-25.34	6
	Lbs TDN/A	10,223.68	11,006.10	-782.42	6
	Lbs NEL/A	10,295.38	11,028.58	-733.20	6
	Lbs DDM/Ton	1,574.90	1,465.23	109.67	6
	LBS CP/Ton	178.40	165.87	12.53	6

<sup>a</sup>Summary over two years

<sup>b</sup>DM=Dry Matter; DDM=Digestible Dry Matter; Tons/A=Tons per acre

<sup>c</sup>ADF=Acid Detergent Fiber; WP Mst=whole plant moisture; UPDM=Whole Plant Dry Matter

<sup>d</sup>TDN=Total Digestible Nutrients in % by weight (dry basis); NEL=Net Energy of Lactation in Mcal per pound.

Table 17

Pairwise Comparison of 330671 and X8311					
	Traits	330671	X8311	Range	#Loc.
<b>Plant Features</b>					
	Plant Hgt	45.28	49.38	-4.10	5
	Ear Hgt	19.06	21.42	-2.36	5
<b>Yield Profile</b>					
	70% Tons/A	27.00	30.23	-3.23	6
	DM Tons/A	8.10	9.07	-0.97	6
	DDM Tons/A	6.22	6.50	-0.28	6
<b>Quality Profile</b>					
	% ADF	23.29	25.39	-2.10	6
	% NDF	44.96	46.97	-2.01	6
	% ASH	4.59	4.32	0.27	6
	% CP	8.49	8.12	0.37	6
	% IVTD	76.65	71.79	4.86	6
	% IVCWD	47.88	39.89	7.99	6
	% WP Mst	69.93	68.17	1.76	6
	% WP DM	30.07	31.83	-1.76	6

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Table 17 (continued)

Pairwise Comparison of 330671 and X8311				
Traits	330671	X8311	Range	#Loc.
<b>Energy Values</b>				
TDN	67.82	65.68	2.14	6
NEL (Mcal/LB)	0.68	0.66	0.02	6
NFC%	38.85	37.50	1.35	6
Lbs CP/A	1,354.67	1,474.91	-120.24	6
Lbs TDN/A	11,019.01	11,942.15	-923.14	6
Lbs NEL/A	11,097.71	11,935.90	-838.19	6
Lbs DDM/Ton	1,533.00	1,435.80	97.20	6
LBS CP/Ton	169.87	162.33	7.54	6

\*Summary over two years of data at 6 locations

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Table 18  
Description of Lines AR5751bm3, AR5152bm3 and AR5151bm3

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AR5751bm3

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<u>COTYLEDON LEAF</u>		<u>EAR CHARACTERISTICS</u>		
ANTHOCYANIN	PRESENT	SILK COLOR	YELLOW	
LENGTH	2-3	EAR LEAVES	ABSENT	
		HUSK		
<u>PLANT AND STALK</u>		COVERAGE	LONG	
PLANT HEIGHT	5.5	LOOSENESS	N/A	
EAR HEIGHT	1.5			
UNIFORMITY RATING	7	EAR ANGLE RATING	7	
ANTHOCY. IN BRACE ROOTS	PRESENT	SHANK LENGTH	4-12	
ANTHOCY. IN NODES	ABSENT	EAR LENGTH	<6	
DIAM. AT 2nd NODE	1.2	EAR SHAPE	CYLINDRICAL/CONICAL	
ROOT RATING	EXCELLENT	UNIFORMITY	7	
20		<u>LEAVES</u>	<u>KERNEL</u>	
ANGLE	UPRIGHT	NO. ROWS	16	
COLOR	LIGHT GREEN	TYPE	DENT	
TOTAL NUMBER	10-15	SIZE	SMALL	
NUMBER ABOVE EAR	5-6	BODY COLOR	ORANGE	
LENGTH, EAR LEAF	26	CROWN COLOR	YELLOW	
MAX., WIDTH, EAR LEAF	3.00			
ANTHOCYANIN, MARGIN	ABSENT			
ANTHOCYANIN, SHEATH	ABSENT	<u>COB</u>		
PUBESCENCE, SHEATH	ABSENT	COLOR	RED	
PUBESCENCE, MARGINS	ABSENT	DIAMETER MIDPOINT	1-2	
PLANT, TILLERING TASSEL	0.00			
35		<u>TASSEL</u>	<u>INSECT RESISTANCE</u>	
COMPACTNESS	AVERAGE	ATTRACTIVE TO APHIDS	NO	
BRANCH ANGLE	30-60			
NUMBER PRIMARY BRANCHES	>6			
SECONDARY BRANCH	ABSENT			
LENGTH	10-14			
40				
TASSEL FERTILITY	1			
SIZE RATING	7			
TASSEL EXTENSION	PARTIALLY ENCLOSED			
SHED IN BOOT?	NO			
45				
DIFFICULTY IN PULLING	AVERAGE			
NUMBER LEAVES PULLED	2			
ANTHER COLOR	YELLOW			
GLUME COLOR - TIP	LIGHT GREEN			
- BASE	GREEN			
- BAND	DARK GREEN			
50				
POLLEN SHED DURATION	N/A			
POLLEN AMOUNT RATING	HEAVY			

55

Table 18 Cont'd

5	<u>AR5152bm3</u>	
	<u>COTYLEDON LEAF</u>	<u>EAR CHARACTERISTICS</u>
	ANTHOCYANIN LENGTH	ABSENT 2-3
10	<u>PLANT AND STALK</u>	<u>HUSK</u>
	PLANT HEIGHT	6.5
	EAR HEIGHT	2.5
	UNIFORMITY RATING	8
15	ANTHOCY. IN BRACE ROOTS	PRESENT
	ANTHOCY. IN NODES	ABSENT
	DIAM. AT 2nd NODE	1.3
	ROOT RATING	EXCELLENT
20	<u>LEAVES</u>	<u>KERNEL</u>
	ANGLE	UPRIGHT
	COLOR	MEDIUM
	TOTAL NUMBER	10-15
	NUMBER ABOVE EAR	5-6
25	LENGTH, EAR LEAF	26
	MAX., WIDTH, EAR LEAF	2.50
	ANTHOCYANIN, MARGIN	ABSENT
	ANTHOCYANIN, SHEATH	ABSENT
	PUBESCENCE, SHEATH	ABSENT
30	PUBESCENCE, MARGINS	PRESENT
	PLANT, TILLERING TASSEL	0.00
	<u>TASSEL</u>	<u>INSECT RESISTANCE</u>
35	COMPACTNESS	AVERAGE
	BRANCH ANGLE	30-60
	NUMBER PRIMARY BRANCHES	4-8
	SECONDARY BRANCH	PRESENT
	LENGTH	10-14
40	TASSEL FERTILITY	3
	SIZE RATING	4
	TASSEL EXTENSION	PARTIALLY ENCLOSED
	SHED IN BOOT?	NO
	DIFFICULTY IN PULLING	EASY
45	NUMBER LEAVES PULLED	2
	ANTHER COLOR	YELLOW
	GLUME COLOR - TIP	RED
	- BASE	YELLOW
	- BAND	DARK GREEN
50	POLLEN SHED DURATION	N/A
	POLLEN AMOUNT RATING	LONG

Table 18 Cont'd

<u>AR5151bm3</u>			
5	<u>COTYLEDON LEAF</u>		<u>EAR CHARACTERISTICS</u>
	ANTHOCYANIN LENGTH	PRESENT 2-3	SILK COLOR YELLOW EAR LEAVES ABSENT <u>HUSK</u>
10	<u>PLANT AND STALK</u>		COVERAGE LONG LOOSENESS N/A
	PLANT HEIGHT	6.0	
	EAR HEIGHT	2.0	
	UNIFORMITY RATING	7	EAR ANGLE RATING 7
15	ANTHOCY. IN BRACE ROOTS	DARK	SHANK LENGTH 4-12
	ANTHOCY. IN NODES	ABSENT	EAR LENGTH <6
	DIAM. AT 2nd NODE	1.0	EAR SHAPE CYLINDRICAL
	ROOT RATING	EXCELLENT	UNIFORMITY 8
20	<u>LEAVES</u>		<u>KERNEL</u>
	ANGLE	HORIZONTAL	NO. ROWS 14
	COLOR	LIGHT GREEN	TYPE SEMI DENT
	TOTAL NUMBER	10-15	SIZE AVERAGE
	NUMBER ABOVE EAR	5-6	BODY COLOR ORANGE
25	LENGTH, EAR LEAF	26	CROWN COLOR YELLOW
	MAX., WIDTH, EAR LEAF	2.50	
	ANTHOCYANIN, MARGIN	ABSENT	
	ANTHOCYANIN, SHEATH	ABSENT	<u>COB</u>
	PUBESCENCE, SHEATH	ABSENT	COLOR PINK
30	PUBESCENCE, MARGINS	ABSENT	DIAMETER MIDPOINT <1
	PLANT, TILLERING TASSEL	0.00	
35	<u>TASSEL</u>		<u>INSECT RESISTANCE</u>
	COMPACTNESS	AVERAGE	ATTRACTIVE TO APHIDS NO
	BRANCH ANGLE	30-60	
	NUMBER PRIMARY BRANCHES	4-6	
	SECONDARY BRANCH	ABSENT	
40	LENGTH	10-14	
	TASSEL FERTILITY	1	
	SIZE RATING	4	
	TASSEL EXTENSION	PARTIALLY ENCLOSED	
	SHED IN BOOT?	NO	
	DIFFICULTY IN PULLING	AVERAGE	
45	NUMBER LEAVES PULLED	1	
	ANTHER COLOR	YELLOW	
	GLUME COLOR - TIP	RED	
	- BASE	LIGHT GREEN	
	- BAND	DARK GREEN	
50	POLLEN SHED DURATION	N/A	
	POLLEN AMOUNT RATING	AVERAGE	

Table 19

AR5751BM3		
Slk Heat 1		1,441

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Table 19 (continued)

AR5751BM3	
5	Slk Heat M 1,463
	Slk Heat F 1,505
	Slk Days 1 81
	Slk Days M 82
	Slk Days F 84
10	Pol Heat 1 1,463
	Pol Heat M 1,485
	Pol Heat F 1,505
	Pol Days 1 82
	Pol Days M 83
15	Pol Days F 84
AR5152BM3	
20	Slk Heat 1 1,587
	Slk Heat M 1,607
	Slk Heat F 1,633
	Slk Days 1 87
	Slk Days M 88
	Slk Days F 89
25	Pol Heat 1 1,534
	Pol Heat M 1,564
	Pol Heat F 1,587
	Pol Days 1 85
	Pol Days M 86
30	Pol Days F 87
AR5151BM3	
35	Slk Heat 1 1,256
	Slk Heat M 1,300
	Slk Heat F 1,345
	Slk Days 1 73
	Slk Days M 75
	Slk Days F 77
40	Pol Heat 1 1,175
	Pol Heat M 1,231
	Pol Heat F 1,345
	Pol Days 1 70
	Pol Days M 72
45	Pol Days F 77

Table 20

Description of Line AR5654bm3	
<b>AR5654bm3</b>	
<u>COTYLEDON LEAF</u>	
ANTHOCYANIN LENGTH	PRESENT 2-3
<u>PLANT AND STALK</u>	
PLANT HEIGHT	6.0
<u>EAR CHARACTERISTICS</u>	
SILK COLOR	PINK
EAR LEAVES	ABSENT
HUSK	
COVERAGE	
LOOSENESS	
COVERS TIP	
N/A	

Table 20 (continued)

Description of Line AR5654bm3				
<b>AR5654bm3</b>				
5	EAR HEIGHT	1.5	EAR ANGLE RATING	6
	UNIFORMITY RATING	7	SHANK LENGTH	4-12
	ANTHOCY. IN BRACE ROOTS	PRESENT	EAR LENGTH	<6
	ANTHOCY. IN NODES	ABSENT	EAR SHAPE	
10	DIAM. AT 2nd NODE	1.1	UNIFORMITY	CYLINDRICAL
	ROOT RATING	EXCELLENT		7
	<u>LEAVES</u>		<u>KERNEL</u>	
	ANGLE	HORIZONTAL	NO. ROWS	14
15	COLOR	MEDIUM	TYPE	FLINT
	TOTAL NUMBER	10-15	SIZE	SMALL
	NUMBER ABOVE EAR	>6	BODY COLOR	ORANGE
	LENGTH, EAR LEAF	19	CROWN COLOR	YELLOW
20	MAX., WIDTH, EAR LEAF	2.00		
	ANTHOCYANIN, MARGIN	ABSENT	<u>COB</u>	
	ANTHOCYANIN, SHEATH	ABSENT	COLOR	RED
	PUBESCENCE, SHEATH	PRESENT	DIAMETER	<1
	PUBESCENCE, MARGINS	ABSENT	MIDPOINT	
25	PLANT, TILLERING TASSEL	0.00		
	<u>TASSEL</u>		<u>INSECT RESISTANCE</u>	
30	COMPACTNESS	LOOSE	ATTRACTIVE TO APHIDS	NO
	BRANCH ANGLE	>60		
	NUMBER PRIMARY BRANCHES	>8		
	SECONDARY BRANCH	ABSENT		
	LENGTH	>14		
35	TASSEL FERTILITY	1		
	SIZE RATING	6		
	TASSEL EXTENSION	PARTIALLY ENCLOSED		
	SHED IN BOOT?	N/A		
40	DIFFICULTY IN PULLING	EASY		
	NUMBER LEAVES PULLED	2		
	ANTHER COLOR	YELLOW		
	GLUME COLOR - TIP	RED		
	- BASE	LIGHT GREEN		
45	- BAND	DARK GREEN		
	POLLEN SHED DURATION	N/A		
	POLLEN AMOUNT RATING	AVERAGE		

Table 21

Description of Line AR5253bm3				
<b>AR5253bm3</b>				
55	<u>COTYLEDON LEAF</u>	PRESENT	<u>EAR CHARACTERISTICS</u>	
	ANTHOCYANIN	2-3	SILK COLOR	YELLOW
	LENGTH		EAR LEAVES	ABSENT
			HUSK	

Table 21 (continued)

Description of Line AR5253bm3				
<b>AR5253bm3</b>				
5	<u>PLANT AND STALK</u>		COVERAGE LOOSENESS	COVERS TIP N/A
	PLANT HEIGHT	6.0		
	EAR HEIGHT	2.5		
	UNIFORMITY RATING	5	EAR ANGLE RATING	6
10	ANTHOCY. IN BRACE ROOTS	PRESENT	SHANK LENGTH	4-12
	ANTHOCY. IN NODES	ABSENT	EAR LENGTH	6-9
	DIAM. AT 2nd NODE	1.1	EAR SHAPE	
	ROOT RATING	EXCELLENT	UNIFORMITY	CYLINDRICAL 8
15	<u>LEAVES</u>		<u>KERNEL</u>	
	ANGLE	UPRIGHT	NO. ROWS	14
	COLOR	MEDIUM	TYPE	SEMI DENT
	TOTAL NUMBER	10-15	SIZE	AVERAGE
20	NUMBER ABOVE EAR	5-6	BODY COLOR	ORANGE
	LENGTH, EAR LEAF	25	CROWN COLOR	YELLOW
	MAX., WIDTH, EAR LEAF	3.00		
	ANTHOCYANIN, MARGIN	ABSENT	<u>COB</u>	
	ANTHOCYANIN, SHEATH	PRESENT	COLOR	
25	PUBESCENCE, SHEATH	PRESENT	DIAMETER	RED
	PUBESCENCE, MARGINS	ABSENT	MIDPOINT	1-2
	PLANT, TILLERING TASSEL	0.00		
30	<u>TASSEL</u>		<u>INSECT RESISTANCE</u>	
	COMPACTNESS	AVERAGE	ATTRACTIVE TO APHIDS	NO
	BRANCH ANGLE	30-60		
	NUMBER PRIMARY BRANCHES	<4		
35	SECONDARY BRANCH LENGTH	PRESENT		
	TASSEL FERTILITY	10-14		
	SIZE RATING	3		
	TASSEL EXTENSION	4		
40	SHED IN BOOT?	PARTIALLY ENCLOSED		
	DIFFICULTY IN PULLING	NO		
	NUMBER LEAVES PULLED	EASY		
	ANTHER COLOR	1		
45	GLUME COLOR - TIP - BASE - BAND	YELLOW RED YELLOW DARK GREEN		
	POLLEN SHED DURATION	N/A		
	POLLEN AMOUNT RATING	AVERAGE		

Table 22

Description of Line AR5153bm3			
<b>AR5153bm3</b>			
55	<u>COTYLEDON LEAF</u>	PRESENT	<u>EAR CHARACTERISTICS</u>
	ANTHOCYANIN		SILK COLOR YELLOW

Table 22 (continued)

Description of Line AR5153bm3				
AR5153bm3				
5	LENGTH	2-3	EAR LEAVES <u>HUSK</u>	ABSENT
	<u>PLANT AND STALK</u>		COVERAGE LOOSENESS	LONG N/A
10	PLANT HEIGHT	6.0		
	EAR HEIGHT	2.0		
	UNIFORMITY RATING	6	EAR ANGLE RATING	7
	ANTHOCY. IN BRACE ROOTS	PRESENT	SHANK LENGTH	<4
	ANTHOCY. IN NODES	ABSENT	EAR LENGTH	N/A
15	DIAM. AT 2nd NODE	1.3	EAR SHAPE	N/A
	ROOT RATING	EXCELLENT	UNIFORMITY	0
	<u>LEAVES</u>		<u>KERNEL</u>	
20	ANGLE	UPRIGHT	NO. ROWS	N/A
	COLOR	MEDIUM	TYPE	N/A
	TOTAL NUMBER	10-15	SIZE	N/A
	NUMBER ABOVE EAR	5-6	BODY COLOR	N/A
	LENGTH, EAR LEAF	28	CROWN COLOR	N/A
25	MAX., WIDTH, EAR LEAF	3.50		
	ANTHOCYANIN, MARGIN	ABSENT	<u>COB</u>	
	ANTHOCYANIN, SHEATH	ABSENT	COLOR	N/A
	PUBESCENCE, SHEATH	ABSENT	DIAMETER	N/A
	PUBESCENCE, MARGINS	PRESENT		
30	PLANT, TILLERING TASSEL	0.00	MIDPOINT	
	<u>TASSEL</u>		<u>INSECT RESISTANCE</u>	
35	COMPACTNESS	COMPACT	ATTRACTIVE TO APHIDS	NO
	BRANCH ANGLE	<30		
	NUMBER PRIMARY BRANCHES	4-6		
	SECONDARY BRANCH	PRESENT		
	LENGTH	10-14		
40	TASSEL FERTILITY	3		
	SIZE RATING	0		
	TASSEL EXTENSION	PARTIALLY ENCLOSED		
	SHED IN BOOT?	YES		
45	DIFFICULTY IN PULLING	EASY		
	NUMBER LEAVES PULLED	2		
	ANTHER COLOR	YELLOW		
	GLUME COLOR - TIP	LIGHT GREEN		
	- BASE	LIGHT GREEN		
50	- BAND	DARK GREEN		
	POLLEN SHED DURATION	N/A		
	POLLEN AMOUNT RATING	LONG		

Table 23

Description of Line AR5551bm3				
<u>AR5551bm3</u>				
5	<u>COTYLEDON LEAF</u>		<u>EAR CHARACTERISTICS</u>	
	ANTHOCYANIN LENGTH	PRESENT 2-3	SILK COLOR EAR LEAVES <u>HUSK</u>	YELLOW LONG
10	<u>PLANT AND STALK</u>		COVERAGE LOOSENESS	SHORT N/A
	PLANT HEIGHT	5.5		
	EAR HEIGHT	1.5		
	UNIFORMITY RATING	7	EAR ANGLE RATING	5
15	ANTHOCY. IN BRACE ROOTS	PRESENT	SHANK LENGTH	>12
	ANTHOCY. IN NODES	ABSENT	EAR LENGTH	6-9
	DIAM. AT 2nd NODE	2.9	EAR SHAPE	CYLINDRICAL
	ROOT RATING	EXCELLENT	UNIFORMITY	5
20	<u>LEAVES</u>		<u>KERNEL</u>	
	ANGLE	DROOPING	NO. ROWS	16
	COLOR	MEDIUM	TYPE	SEMI DENT
	TOTAL NUMBER	10-15	SIZE	AVERAGE
25	NUMBER ABOVE EAR	5-6	BODY COLOR	ORANGE
	LENGTH, EAR LEAF	30	CROWN COLOR	YELLOW
	MAX., WIDTH, EAR LEAF	4.00		
	ANTHOCYANIN, MARGIN	ABSENT	<u>COB</u>	
30	ANTHOCYANIN, SHEATH	ABSENT	COLOR	WHITE
	PUBESCENCE, SHEATH	ABSENT	DIAMETER	1-2
	PUBESCENCE, MARGINS	PRESENT	MIDPOINT	
	PLANT, TILLERING TASSEL	0.00		
35	<u>TASSEL</u>		<u>INSECT RESISTANCE</u>	
	COMPACTNESS	LOOSE	ATTRACTIVE TO APHIDS	NO
	BRANCH ANGLE	>60		
	NUMBER PRIMARY BRANCHES	>8		
40	SECONDARY BRANCH	ABSENT		
	LENGTH	>14		
	TASSEL FERTILITY	1		
	SIZE RATING	6		
45	TASSEL EXTENSION	PARTIALLY ENCLOSED		
	SHED IN BOOT?	NO		
	DIFFICULTY IN PULLING	AVERAGE		
	NUMBER LEAVES PULLED	1		
	ANTHER COLOR	YELLOW		
50	GLUME COLOR - TIP	LIGHT GREEN		
	- BASE	LIGHT GREEN		
	- BAND	DARK GREEN		
	POLLEN SHED DURATION	N/A		
55	POLLEN AMOUNT RATING	HEAVY		

Eleven *bm* hybrids were developed from inbreds homozygous for *bm3*. These hybrids were grown and ensiled at six U.S. locations for 2 years and pairwise comparisons were made between each *bm* hybrid and the corresponding

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normal isogenic hybrid. The results for forage yield, IVTD and *in vitro* NDF digestibility are summarized in Table 24. Hybrids are listed in Table 24 according to their maturity rating. For example, hybrid number 330659 is a short season (95 day MMR) type, whereas 330670, 330671 and 330672 are long season (115 day MMR) types.

5 The results indicate that  $F_1$  *bm* hybrids can be developed whose silage has an NDF digestibility of 47% or greater. The results also show that *bm* hybrids can be developed and adapted that have an IVTD of 75% or greater and a forage yield that is only 3-15% less than the yield of the isogenic normal hybrid. For example, hybrid 330662 has a 105 day maturity rating and an average NDF dig. of 51.78%, yet has a forage yield of 25.59 tons/acre. Hybrid 330671 has a 115 day maturity rating and an average NDF dig. of 47.88%, yet has a forage yield of 27.00 tons/acre. Moreover, these results indicate that *bm* hybrids derived from different heterotic groups and adapted to different maturities can 10 be developed. Silage from such *bm* hybrids is suitable for feeding to cattle as disclosed herein.

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Table 24

Pairwise Comparisons of Silage from 13 bm Hybrids and Isogenic Normal Hybrids *C*

Hybrid No.	% CP	MTD(%)		% NDF		Forage Yield (Tons/Acre at 70% Moisture)		Inbred Parents for bm Hybrid <i>e</i>	
		bm <sup>a</sup>	Normal	bm <sup>a</sup>	Normal	bm <sup>a</sup>	Normal	bm <sup>a</sup>	Normal
330659	8.51	8.23	77.06*	74.50	49.80*	40.88	45.48	43.14	20.13*
330663	8.95*	7.83	78.03*	73.05	50.89*	38.20	44.81	43.52	22.35
330661	7.46	7.61	77.32	73.90	48.35	42.32	43.88	45.22	23.72
330684	7.86*	7.26	76.88*	74.04	47.99*	41.37	44.36	44.44	23.21*
330662	8.20	7.72	78.71	76.81	51.78*	42.15	44.07	40.06	25.59
330685	8.55	8.23	78.22*	73.47	50.30*	41.37	45.03	45.25	24.39
330668 <sup>d</sup>	8.92	8.29	78.75*	73.26	50.99*	40.84	43.56	45.08	25.00
330667	8.93	8.22	78.33*	73.76	50.86*	40.28	44.41	44.28	24.76
330669	9.28	8.62	78.58*	74.33	48.43*	38.08	41.54	41.80	24.87*
330668	8.87	8.74	78.28*	73.86	49.79*	38.87	43.52	42.82	25.13
330670	8.13*	7.32	78.86*	71.91	48.35*	41.02	45.27	47.89	25.72
330671 <sup>d</sup>	8.49	8.12	76.65*	71.79	47.88*	39.89	44.98	46.97	27.00
330672	8.24	8.28	76.41*	71.77	49.85*	40.54	47.27	47.84	25.33*

<sup>a</sup>Values with an asterisk are significantly different from isogenic normal silage at p<0.05 (Student's t-test)<sup>b</sup>Percent decrease in forage yield of bm silage relative to isogenic normal silage<sup>c</sup>Summary over two years at 5 or 6 locations<sup>d</sup>Data for hybrid nos. 330686 & 330671 are also presented in Tables 16 and 17, respectively<sup>e</sup>Corresponding isogenic normal inbreds were used to make the normal hybrids

Example 5**Formulation of a Dairy Cattle Feed Ration**

5 Control and *bm* F<sub>1</sub> hybrid seed of Example 1 was planted and cultivated in the Midwest United States using a standard agronomic practices. The corn was harvested approximately 5 months after planting, using standard corn silage harvesting methods. The harvested material was ensiled in 150T capacity concrete bunker silos. The harvested material was not treated with inoculant or preservative.

10 A subsample of each load was ensiled in a 10.16 centimeter (cm) diameter X 40.64 cm length PVC experimental silo fermentation canister made of polyvinyl chloride. Each silo contained a 0.64 cm X 2.54 cm piece of copper tubing inserted into the sidewall. The tubing was covered with a rubber policeman having a slit that allowed fermentation gases to escape while preventing entry of ambient air. Each fresh-cut silage sample was packed tightly into a silo and allowed to ferment for 30 days at ambient temperatures (above freezing) before opening and assaying for nutrient concentrations.

15 The results of the nutrient concentration assays are shown in Table 25. *In vitro* NDF digestibility was measured as described in Goering, H. and Van Soest, P., *supra*, except that the fermentation period was 30 hours instead of 48 hours. The control corn silage and the BMR corn silage were similar in crude protein and NDF and differed in 30 hour *in vitro* NDF digestibility.

Table 25

Nutrient composition of corn silages <sup>1</sup>		
Nutrient	Control	BMR
Dry matter, % as fed	33.5	30.2
Crude protein, %	8.35	8.68
ADF, %	21.02	21.08
NDF, %	40.38	42.02
Hemicellulose, %	19.36	20.94
NDF dig., %	36.8	45.3
Ash, %	3.76	4.24

<sup>1</sup>Nutrients and digestibility are expressed on a dry matter basis.

35 Experimental diets were developed to be fed as a total mixed ration (TMR), using the following assumptions: the early lactation experimental dairy cow was estimated to weigh 613.6 kg and to produce 45.45 kg of milk per day with 3.5% fat, 3.30% milk protein and a 0.18 kg/day increase in body weight. Dry matter intake (DMI) was predicted to be slightly less than 4.0% of body weight.

40 The two TMRs were formulated to contain 56% forage on a dry matter basis. The forage consisted of 4 parts corn silage (either normal or BMR) to 1 part alfalfa haylage on a dry matter basis. The TMR also contained soybean meal (SBM, 44% crude protein), high moisture corn, finely ground dry corn, whole linted cottonseeds, mineral and vitamins. The two diets were balanced to be isonitrogenous and varied only in the proportions of SBM and high moisture corn. Rations contained approximately 18.0% crude protein, 44% of which was soluble protein and 31% of which was rumen 45 undegraded protein (rumen bypass protein). Both rations were balanced to meet or exceed mineral and vitamin requirements. The ingredient composition of the two rations is given in Table 26. The nutrient composition of the two rations is given in Table 27.

Table 26

Ingredient Composition of Total Mixed Ration <sup>1</sup>		
INGREDIENT	Normal Silage (% by weight)	BMR Silage (% by weight)
Corn silage	44.6	44.6
Alfalfa silage	11.2	11.2
Soybean Meal (44% protein)	19.2	18.8

<sup>1</sup>Dry matter basis.

Table 26 (continued)

Ingredient Composition of Total Mixed Ration <sup>1</sup>		
INGREDIENT	Normal Silage (% by weight)	BMR Silage (% by weight)
Corn, high moisture	8.8	9.2
Corn, dry ground	5.6	5.6
Cottonseeds, whole linted	5.6	5.6
Minerals and Vitamins	5.0	5.0
Total	100.0	100.0

<sup>1</sup>Dry matter basis.

Table 27

Nutrient Composition of Total Mixed Ration <sup>1</sup>		
NUTRIENT	Normal Silage	BMR Silage
Dry Matter	46.8	43.8
Crude Protein (CP)	18.0	18.0
Soluble Protein(SP as % of CP)	43.8	44.2
RUP (RUP as % of CP)	31.0	31.0
Fat	3.8	3.8
Net Energy of Lactation (NE <sub>L</sub> ) <sup>2</sup> Mcal/kg	1.72	1.72
Acid Detergent Fiber (ADF)	18.4	18.5
Neutral Detergent Fiber(NDF)	30.6	31.3
Non-Fiber Carbohydrate (NFC)	39.6	38.8
Calcium	1.01	1.01
Phosphorus	0.45	0.45
Magnesium	0.29	0.29
Sulfur	0.22	0.22
Salt	0.76	0.76

<sup>1</sup>Percentages for nutrients given as % by weight on a dry matter basis. Dry matter is given as % by weight of ration as fed.

<sup>2</sup>Book values (1.67 Mcal/kg) used for corn silage.

### Example 6

#### **Milk Production Using BMR Silage In a Dairy Cattle Ration**

32 lactating dairy cows were utilized in a complete block design experiment, consisting of 2 primiparous cows and 14 multiparous cows fed a ration containing control silage and 2 primiparous cows and 14 multiparous cows fed BMR silage. Cows within blocks were randomly assigned to treatments. All cows were disease free and otherwise healthy. Rations were formulated as described in Example 5.

The experiment was a crossover design with 28 day periods. The first 21 days of the period were used for diet adjustment and the last 7 days were used for data collection. Cows ranged from approximately 22 to 141 days in milk, averaging 90 days in milk at the beginning of the experiment.

Cows were milked 3 times daily. Milk production was measured at each milking and reported daily. Nine samples of milk were taken on 3 different days from each cow for composition analysis in each collection period. Composition analysis included fat, protein, lactose, and somatic cell counts. Composition analysis was carried out using Near Infrared Spectroscopy at the Michigan Dairy Herd Improvement Association, East Lansing, MI. Body weights were recorded on 2 consecutive days before the experiment started and on the last 2 days of each data collection period.

Body condition scores were recorded on the day before the experiment started and on the last day of each data collection period. Scores were recorded according to the definitions indicated in Table 6. Three individuals recorded condition scores at each measurement time.

5 After 28 days, cows were abruptly switched from one ration to the other ration (from ration containing BMR silage to ration containing control silage and vice versa). Cows were fed and milked in the same manner as prior to the switch in ration. Milk was weighed and analyzed in the same manner as prior to the switch in ration. The experiment was terminated after 56 days. Data were analyzed using the fit model procedure in the JMP computer statistics program (SAS Institute, North Carolina).

10 The milk production results are shown in Table 28. The results indicate that cows fed BMR silage have a statistically significant increase in milk production when comparing treatment means. The increase was more than 2.7 kg of milk/cow/day on an uncorrected basis and about 2.6 kg milk/cow/day on a solids-corrected basis (Table 28, lines 2 and 5). There was no significant change in the percentage milk fat, protein, lactose, or solids-not-fat (SNF) between milk from cows fed control or BMR silage (Table 28, lines 6-9).

15 Dry matter intake (DMI) by cows fed BMR silage was significantly greater than that of cows fed control silage (Table 28, line 1). It is noteworthy that there was no significant difference in mean milk production for the two rations when dry matter intake (DMI) was used a covariate ( $P=0.42$ ). This result suggests that increased milk production is due to an increase in the amount of total ration consumed per day when BMR silage was fed, i.e., that the rate of intake and feed conversion was increased.

20 Because milk from BMR silage-fed cows has a composition similar to that of milk from control silage-fed cows, milk from BMR-fed cows can be pooled with milk from other sources and processed by standard techniques. Moreover, milk from BMR silage-fed cows typically will not be subject to a price penalty due to non-standard component concentrations.

25 The effect of a ration comprising BMR silage on body condition and weight is shown in Table 29. Feeding of such a ration resulted in a greater weight gain than did feeding of a ration comprising control silage, although this difference was not statistically significant. Moreover, the body condition score change was significantly better when cows were fed BMR silage than when cows were fed control silage ( $P=0.049$ ; Table 29). These results indicate that not only does feeding of a ration comprising BMR silage as disclosed herein increase milk production, cows fed such a ration have improved body condition scores. Improved body condition scores are a general measure of a cow's ability to sustain milk production over an extended period of time, e.g., multiple lactations.

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Table 28

Milk Production				
	Component	Control Silage	BMR Silage	P Value
35	Dry Matter Intake (DMI) <sup>e</sup>	23.5	25.5	<0.0001
	Milk Yield <sup>c</sup>	39.0	41.7	0.002
40	4% FCM <sup>a</sup>	35.7	38.0	0.0005
	3.5% FCM <sup>f</sup>	38.5	41.0	0.0005
	Solids Corrected Milk <sup>g</sup>	35.2	37.8	0.0001
45	Milk Fat (%) <sup>d</sup>	3.46	3.43	0.71
	Milk Protein (%)	2.95	2.99	0.35
	Milk Lactose (%)	4.85	4.90	0.22
50	SNF <sup>b</sup> (%)	8.52	8.59	0.13

<sup>b</sup> = Percentage of Solids-not-fat.

<sup>c</sup> = Milk production is expressed as kg/cow/day, unless indicated otherwise.

<sup>d</sup> = Dry matter intake is expressed a kg per cow per day.

<sup>e</sup> = Milk components are expressed as a percentage (gm/100ml of milk), unless indicated otherwise.

<sup>f</sup> = Milk yield on a fat-corrected basis to 3.5% milkfat. (Milk x 0.4255)-(16.425 x Fat content/100 x milk).

<sup>g</sup> = Tyrrell, H.F. and J.J. Reid, J. Dairy Sci. 48:1215 (1965).

<sup>b</sup> = Milk yield on a fat-corrected basis to 3.5% milkfat. (Milk x 0.4255)-(16.425 x Fat content/100 x milk).

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Table 28 (continued)

Milk Production			
Component	Control Silage	BMR Silage	P Value
Milk Fat (kg/cow/day)	1.33	1.42	0.01
Milk Protein (kg/cow/day)	1.14	1.24	0.0001
Milk Lactose (kg/cow/day)	1.88	2.04	0.0005

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Table 29

Effect of BMR Silage on Body Condition and Weight Change			
Component	Control Silage	BMR Silage	P Value
Body Weight Change (kg increase after 28 days)	2.2	4.76	0.45
Body Condition Score Change after 28 days	0.019	0.110	0.049
Ratio: 4% FCM/DMI	1.52	1.48	0.23

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Example 7**Milk Production Using a BMR Silage Ration and Bovine Somatotropin**

25 An on-farm herd of about 80 lactating dairy cows was utilized to determine the effect of feeding BMR silage in combination with bovine somatotropin (BST) treatment. Cows were fed in a switchback trial, consisting of feeding a control ration for the first phase, feeding a BMR ration for the second phase, followed by a return to a control ration.

15 BMR corn hybrid 330666 (Table 24), produced from a cross of inbreds AR5252bm3 X 7675bm3, was grown and harvested for silage at the half milk line stage, using typical commercial farm practices. The material was ensiled for 30 about 75 days in an outdoor environment before feeding. The nutrient composition of the silage was similar to that shown in Table 24. Control silage from a locally adapted normal hybrid was grown and harvested in the same manner.

20 Experimental diets were developed to be fed as a total mixed ration (TMR), assuming a dairy cow 100 days in milk, weighing 613.6 kg and producing 45.45 kg of milk per day with 3.9% fat, 3.30% milk protein. No increase in body weight was predicted. Dry matter intake (DMI) was predicted to be slightly greater than 4.0% of body weight.

25 The TMRs were formulated to contain about 46-47% forage on a dry matter basis. The forage consisted of approximately equal parts corn silage (normal or BMR) and alfalfa haylage on a dry matter basis. The TMRs also contained heated soybean flakes, high moisture corn, whole linted cottonseeds, and Gro-Mark(tm) Pro-Barley Mix, a proprietary mixture believed to comprise grain, minerals and vitamins. The TMRs were balanced to be isonitrogenous and to meet or exceed mineral and vitamin requirements. Rations contained slightly more than 18% crude protein, about 31-32% 40 of which was soluble protein. The ingredient composition and the nutrient composition of the total mixed rations is given in Tables 30-31.

30 About 75% of the cows in the herd were treated with BST during the trial. BST was not administered to cows less than 30 days in milk nor to cows not confirmed to be pregnant; such cows constituted about 25% of the herd. BST (Posilac(tm), Monsanto, St. Louis, MO) was administered according to the manufacturer's instructions, using dosages 45 recommended by the manufacturer.

Table 30

Ingredient Composition of Total Mixed Ration <sup>1</sup>			
INGREDIENT	TMR <sup>2</sup>		
	Days 1-10 (Control Silage)	Days 11-21 (BMR Silage)	Days 22-31 (Control Silage)
Corn silage	22.57	21.52	22.16
Alfalfa silage	24.90	25.79	23.76

1 Dry matter basis.

2% by weight.

Table 30 (continued)

INGREDIENT		TMR <sup>2</sup>		
		Days 1-10 (Control Silage)	Days 11-21 (BMR Silage)	Days 22-31 (Control Silage)
Soybean Flakes, heated		2.60	3.00	3.94
Corn, high moisture		25.05	24.92	25.36
Cottonseeds, whole linted		4.60	4.59	4.66
Gro-Mark <sup>tm</sup> ProBarley Mix		20.28	20.18	20.13
Total		100.0	100.0	100.0

<sup>1</sup>Dry matter basis.<sup>2</sup>% by weight.

Table 31

NUTRIENT		TMR		
		Days 1-10	Days 11-21	Days 22-31
Dry Matter		51.62	48.58	55.41
Crude Protein (CP)		18.58	18.53	18.16
Soluble Protein(SP as % of CP)		32.42	32.05	30.82
Crude Fat		5.59	5.56	5.83
Net Energy of Lactation (NE <sub>L</sub> ) <sup>2</sup> Mcal/kg		1.72	1.72	1.74
Acid Detergent Fiber (ADF)		18.87	18.70	18.50
Neutral Detergent Fiber(NDF)		29.43	28.92	28.81
Calcium		1.08	1.10	1.05
Phosphorus		0.57	0.57	0.56
Magnesium		0.34	0.35	0.34
Sulfur		0.26	0.26	0.26
Salt		0.56	0.55	0.55

<sup>1</sup>Percentages for nutrients given as % by weight on a dry matter basis. Dry matter is given as % by weight of ration as fed.<sup>2</sup>Book values (1.67 Mcal/kg) used for corn silage.

Cows ranged from approximately 7 to 489 days in milk, averaging about 211 days in milk at the beginning of the experiment. Cows were milked twice daily. Milk production for the herd was measured at each milking. Cows were fed and milked in the same manner throughout the experiment. On day 1 of the trial, selected cows were treated with BST and the herd was placed on control ration (containing control silage). The herd was abruptly switched from control ration to BMR ration (containing BMR silage) on day 11. On the same day, selected cows were injected with BST. At day 22, cows were abruptly returned to the control ration. At day 25, selected cows were injected with BST.

Herd milk production results are shown in Table 32 and presented graphically in Figure 1. When the herd was fed a control feed ration, average daily milk production of the herd began to increase following treatment of selected cows with BST. Daily milk production peaked about 5-6 days at about 39 kg/cow/day and then declined to about 36 kg/cow/day. A peak in milk production between injections is typical of prolonged release BST administration.

The herd was switched to a ration comprising BMR silage at day 11. In the second phase of the trial, average milk production increased to about 39-40 kg/cow/day at 6 days after the BST treatment and ration change. Milk production

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remained at about 39-40 kg/cow/day for the duration of this phase of the trial. These results show that delivery of BST to cows in conjunction with the feeding of a ration comprising BMR silage results in a sustained increase in average daily milk production rather than the decline in milk production typically observed when BST treatment alone is used.

5 The sustained increase in milk production during the BMR ration phase is also shown by comparing the average daily milk production for the last four days of each phase. Average milk production for the last 4 days of the first phase (control ration/BST) was 37.50 kg of milk/cow/day on an uncorrected basis. Average milk production for the last 4 days of the second phase (BMR ration/BST) was 40.45 kg of milk/cow/day on an uncorrected basis.

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Table 32  
Milk Production With BMR and BST

5	Diet	Trial Day	Cows		TMR Mix (kg)		Milk Production (kg)	
			Tested	Milked	Morn	Eve	Tank Weight	Avg/Cow
10	Control	1 <sup>a</sup>	73	69	1636	1636	2568	37.22
		2	73	69	1636	1636	2566	37.18
		3 <sup>b</sup>	71	66	1636	1636	2580	39.10
		4	71	66	1636	1636	2556	38.73
		5	71	66	1636	1636	2612	39.57
		6 <sup>c</sup>	70	67	1636	1636	2670	39.86
20		7	70	67	1636	1636	2600	38.80
		8	70	67	1636	1636	2556	38.15
		9 <sup>d</sup>	69	66	1636	1636	2444	37.02
		10 <sup>e</sup>	70	67	1636	-- <sup>f</sup>	2433	36.31
30	BMR	11 <sup>g</sup>	72	67	1818	1818	2357	35.18
		12 <sup>h</sup>	73	70	1818	1750	2422	34.60
		13	73	70	1818	1750	2560	36.58
		14	73	70	1750	1750	2587	36.95
		15	73	70	1818	1818	2666	38.09
		16 <sup>i</sup>	73	71	1818	1886	2682	37.77
40		17	73	71	1841	1832	2796	39.39
		18 <sup>j</sup>	71	69	1836	1818	2774	40.20
		19	71	69	1818	1827	2836	41.10
		20 <sup>k</sup>	70	68	1809	1823	2763	40.64
		21	70	68	1827	1850	2710	39.84
50	Control	22	70	68	1682	1673	2682	39.44
		23	70	67	1682	1682	2545	37.99
		24	70	67	1682	1682	2500	37.31
		25 <sup>l</sup>	70	67	1591	1609	2450	36.56
		26 <sup>m</sup>	70	67	1600	1591	2381	35.54

Diet	Trial Day	Cows		TMR Mix (kg)		Milk Production (kg)	
	27	70	67	1591	1609	2442	36.45
	28	70	67	1595	1595	2545	37.99
	29	70	67	1559	1550	2585	38.58
10	30 <sup>a</sup>	69	66	1555	1545	2563	38.84
	31	69	66	1568	1550	2642	40.03

<sup>a</sup> = BST Injection; <sup>b</sup> = 2 Dry; 1 Mastitis; <sup>c</sup> = 2 Fresh; 1 Dry; <sup>d</sup> = 1 Died; 1 Sick; <sup>e</sup> = 1 Fresh; <sup>f</sup> = Not Determined; <sup>g</sup> = BST Injection; <sup>h</sup> = 2 Fresh; 2 Off-feed; <sup>i</sup> = 1 Fresh; <sup>j</sup> = 1 Displaced Abomasum (DA) Surgery; 1 Dry; <sup>k</sup> = 1 Dry; <sup>l</sup> = BST Injection; <sup>m</sup> = 1 DA Surgery; <sup>n</sup> = 1 Dry

20 To the extent not already indicated, it will be understood by those of ordinary skill in the art that any one of the various specific embodiments herein described and illustrated may be further modified to incorporate features shown in other of the specific embodiments.

25 The foregoing detailed description has been provided for a better understanding of the invention only and no unnecessary limitation should be understood therefrom as some modifications will be apparent to those skilled in the art without deviating from the spirit and scope of the appended claims.

#### Claims

30 1. A ruminant animal feed comprising a combination of:

35 a forage component comprising from about 20% to about 60% of said feed on a dry matter basis, said forage component comprising from about 20% to about 100% corn silage on a dry matter basis produced from corn plants exhibiting a brown midrib (*bm*) phenotype, said silage having an *in vitro* neutral detergent fibre digestibility of about 44% to about 70%; and  
a feed composition component;  
said animal feed having a fibre content of about 20% to about 40%.

40 2. A feed as claimed in claim 1, wherein said neutral detergent fibre digestibility is from about 6 percent to about 20 percent greater than the neutral detergent fibre digestibility of corn silage produced from corresponding isogenic normal corn plants.

45 3. A feed as claimed in claim 1 or 2, wherein said corn plants exhibiting a (*bm*) phenotype comprise *F*<sub>1</sub> hybrid plants.

50 4. A feed as claimed in any one of claims 1 to 3, wherein said corn plants are homozygous for *bm3*.

55 5. A feed as claimed in any one of claims 1 to 4, wherein said silage has a whole plant *in vitro* digestibility from about 65% to about 85%.

6. A feed as claimed in any one of claims 1 to 5, wherein said animal feed has a crude protein content of from about 17% to about 21% on a dry matter basis and about 35% to about 50% of said crude protein is soluble protein.

7. A method of enhancing milk production in a ruminant animal, comprising the step of feeding said animal an animal feed as defined in any one of claims 1 to 6.

8. A method as claimed in claim 7 wherein said animal is a dairy cow.

9. A method as claimed in claim 7 or 8, further comprising the step of administering a biologically active somatotropin

to said animal under conditions delivering an effective amount of said somatotropin to said animal during a selected period.

10. A method as claimed in claim 9, wherein said somatotropin is administered as a prolonged release dose.
- 5 11. A method as claimed in claim 8 or 9, wherein said conditions comprise delivering said somatotropin to the circulatory system of said animal.
12. A method as claimed in any one of claims 9 to 11, wherein said dose is effective for at least 7 days.
- 10 13. A method of producing a total ruminant animal feed as defined in any one of claims 1 to 6 comprising the steps of:
  - 15 a) obtaining a corn silage produced from corn plants exhibiting a *bm* phenotype; and
  - b) formulating a total ruminant animal feed comprising a combination of:
    - a forage component and
    - a feed composition component as defined in any one of claims 1 to 6.
14. A method as claimed in claim 13 wherein said corn plants are grown from a substantially homogenous assemblage of corn seeds which are homozygous for at least one *bm* allele.
- 20 15. A pack for a ruminant animal feed comprising a forage component and separately a feed composition component as defined in any one of claims 1 to 6 as a combined preparation for simultaneous, separate or sequential use to enhance milk production in said animal.
- 25 16. A pack as claimed in claim 15 additionally comprising a biologically active somatotropin.

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Figure 1

